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PRACTICAL MICROSCOPY

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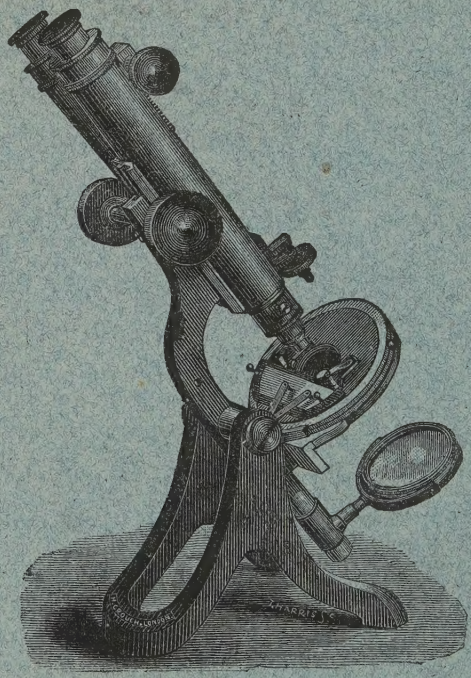

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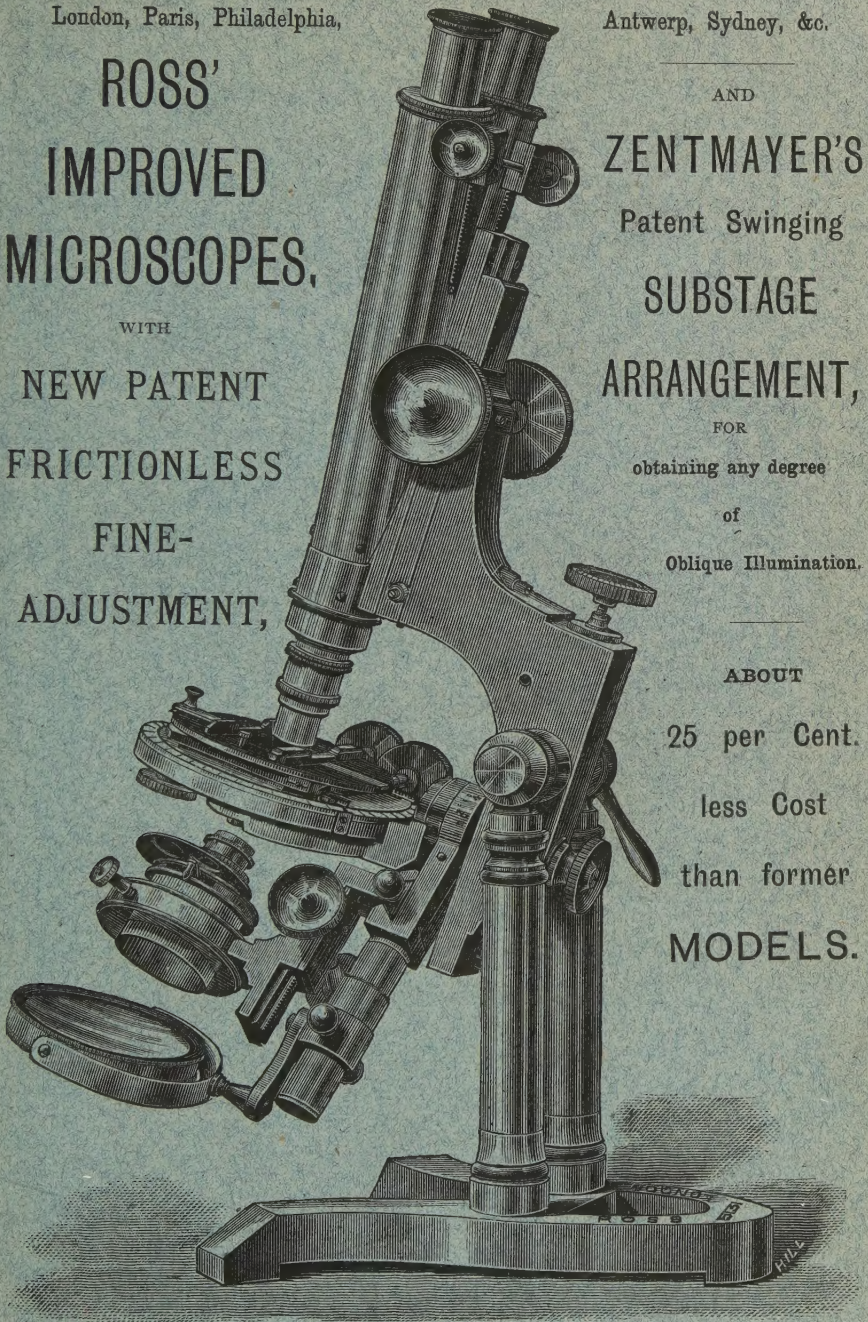

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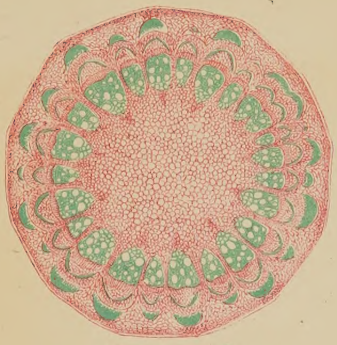

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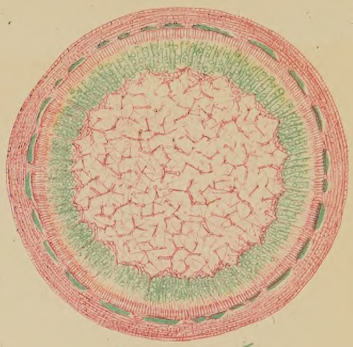

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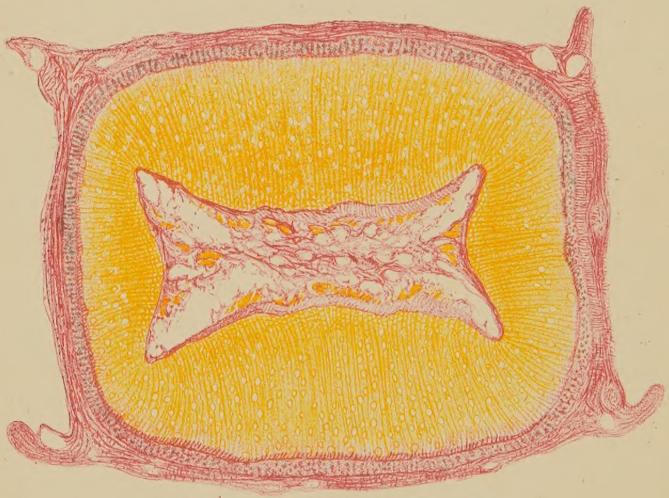

EUCALYPTUS GLOBULUS.

GOUT WEED.

BLACK PEPPER.

WOOD SECTIONS — DOUBLE STAINED.

PRACTICAL MICROSCOPY.

BY

GEORGE E. DAVIS,

F.R.M.S., F.I.C., F.C.S.,

ETC., ETC.

ILLUSTRATED WITH TWO HUNDRED AND FIFTY-SEVEN WOODCUTS AND A COLOURED FRONTISPIECE.

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PREFACE.

THE Author's object in presenting this work to the student of microscopy is to supply at a reasonable cost a book written upon somewhat similar lines to Quekett's 'Practical Treatise on the Use of the Microscope,' the second and last edition of which appeared in 1852. The necessity of a treatise bringing the science to the present day has been long felt, and it is hoped that a vacant place in microscopical literature is now filled up.

Although essentially a practical work upon microscopy, the reader is treated to a little theory, in the hope that it may lead to a more minute study of the optical principles upon which the microscope is constructed. Some may object, perhaps, to so little theory being introduced, especially on those subjects intimately connected with Professor Abbe's recent researches; but it should be remembered that the addition of such matter would have added considerably to the cost of production and detracted somewhat from the practical character of the book: this, taken together with the fact that ample references have been given to Professor Abbe's papers, will, it is hoped, be sufficient apology. Moreover, the information lately put forth is novel to the general body of microscopists; the views now held are so

decidedly opposite to those promulgated but a few years ago, that it is evident any one writing on the subject now could only put forth an abstract of the learned Professor's views. This the Author did not wish to attempt, as such a proceeding would only have mystified the student, and not have furnished the advanced worker with as much information as he could obtain from the 'Journal of the Royal Microscopical Society,' where Professor Abbe's papers may be found *in extenso.*

With regard to the selection of objectives, the Author hopes he will not be misunderstood:—medium angles have been advised for students' use, for the simple reason that they can be employed without much previous knowledge or difficulty; but for all purposes of scientific investigation, wide apertures, requiring often much skill in manipulation, will give the most satisfactory results.

Our American brethren have for many years been impressed with the importance of wide apertures, and no doubt many of their arguments were sound, though it cannot be said that their case was ever well demonstrated and supported.

Dr. G. E. Blackham, in a paper read before the Microscopical Congress at Indianapolis in 1878, argued exclusively for wide apertures, though he was singularly unfortunate in his selection of a theory to account for "penetration" in objectives; while this *brochure* has been supplemented by a volume from the pen of Professor J. E. Smith, 'How to See with the Microscope,' putting forward again the merits of wide angles.

Microscopists who have read these works will do well

to study what Professor Abbe has written, and then they will probably come to the conclusion that the high-angle school possess some forcible arguments, and may also find that with wide apertures their battery of objectives may be considerably smaller to accomplish work of a better quality than when low angles are employed.

Under the head of objectives, the almost total absence of American productions will be remarked: it is only recently that American objectives of the widest aperture have found their way into the Author's hands. Their definition is marvellous, but it has not been thought advisable to include such objectives in a practical work until after they have been used in ordinary observation for a sufficient length of time.

However, the reader may like to know that Spencer, of Geneva, U.S.A., produces a 3-inch of 13°, a 2-inch of 20°, and a 1-inch of 50°; while Tolles, of Boston, U.S.A., makes similar objectives, a $\frac{4}{10}$ of 145°; both of these opticians producing the $\frac{1}{4}$-inch and all higher powers, of 180° air-angle.

These wide apertures demonstrate clearly the accuracy of the statement made on page 51, that wide angles require more care in their correction, and are consequently more expensive. Spencer's 1-inch of 50° costs forty-five dollars, or 9*l.*; while the 1-inch of 22° in their student's series costs but ten dollars, or 2*l.*

The whole of the information in the work has been selected to aid the student as much as possible; but it should be remembered that the microscope is useful, from a scientific point of view, only as an aid in research, unfolding to us objects either invisible, or but faintly to be distinguished

by the unassisted eye. The Author has not pandered to the tastes of mere lovers of pretty objects.

As to the preparation and mounting of objects much more might have been said; indeed, volumes written on this subject alone; but types have been selected to avoid repetition, and which the student will do well to follow. All the processes in practical microscopy must be carried out with intelligence, and the why and the wherefore of each well understood. If this be done the student will find no insuperable difficulties.

Many of the illustrations found herein have been photographed from nature by the Author, and cut in the engraving department of 'Design and Work,' and thanks are here expressed to the proprietor for these blocks, which have enabled the work to be so fully illustrated at a moderate cost. To the publisher of 'Science-Gossip,' for similar reasons, thanks are likewise expressed. Many of the illustrations have been lent from the 'Northern Microscopist.'

In conclusion, the Author asks his readers to communicate to him, under care of the publisher, any ideas or suggestions in view of future editions, and hopes he has established a sufficient *raison d'être.*

GEORGE E. DAVIS.

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PRACTICAL MICROSCOPY.

CHAPTER I.

INTRODUCTION.

DURING the past few years rapid strides have been made in the manufacture and supply of cheap and at the same time really good working microscopes; and these having been extensively purchased, have extended and disseminated a taste for the study of minute things, in those to whom the possession of a microscope a few years ago seemed a luxury to be only dreamt of.

Good works treating upon the practical use of the microscope and its accessories, have hitherto been of so expensive a character as to be certainly beyond the reach of most students, and many who might have been led into the paths of scientific research, have probably kept their instruments to gratify the curiosity of themselves and their friends. Such persons often think that the possession of a microscope must always make a microscopist, and in order to study objects, that it is only necessary to place them on the stage of the microscope when their hidden structure will manifest itself at once. No greater mistake could ever be made. It is very desirable that the student should be thoroughly acquainted with the physics and chemistry of the science, for when once these are mastered

he will see his way to carry on experiments in a rational manner, as often as fresh problems are presented to him.

Again, no inexpensive work upon practical microscopy brings the subject down to the present time; this and other minor details urged us to cut out a path for ourselves, and produce a treatise which might be of use to the student.

In using the microscope as a means of gaining an insight into the anatomy or life-history of forms only faintly distinguished by the unassisted eye, the student must be prepared to exercise a certain amount of patience; skill in manipulation comes by practice only, and before commencing with any investigation, all preconceived notions should be cast aside, the observer being guided chiefly by the results of his own observations.

Isolated observations should never be considered conclusive; experiments require frequent repetition under very varying conditions before we can accept the results as true interpretations of what we have actually seen. We should also be careful to take into account the degree of perfection of our instruments as well as our own capability of vision; what one observer will see, another will fail to detect, and if any preconceived notions exist, the observations are sure to be moulded after the same fashion.

Expertness in microscopy is only to be attained by study and practice, and as it is necessary, in order to progress, that we take up the subject where our predecessors left off, it is necessary to become acquainted with what has been done previously, and this may be arrived at by the perusal of old standard works upon the science, such as those of Baker, Pritchard, and Goring, or the more recent work of Mr. Quekett's, 'The Practical Treatise on the Use of the Microscope.' These works, though old, are valuable, and

the executive of every microscopical society should endeavour to secure a copy of each for its library.

If microscopical students would only peruse these older works, and enter into communication and discussion with their brother microscopists, much time and work would be saved, and a great deal of that *re-discovery* which goes on at the present day averted; there is no other way of avoiding repetitions except by the methods above stated.

We might advance more rapidly, or more certainly at least, if we asked, before performing any microscopical operation, the reason why. We are too apt to do things in a certain manner because some one who instructed us in the art and mystery did so before us; no inquiry was made, but we did likewise. If the principles upon which all operations are conducted were thoroughly understood by all those who intend working with the microscope, much labour would be saved, needless experiments avoided, slides rendered more permanent, and microscopical research brought more into favour on account of some of the barriers which now obstruct progress having been broken down.

It is in this direction that a treatise may be useful to the student; objects themselves, whether of animal, vegetable, or mineral origin, are best treated of in works entirely independent of practical microscopy; an organism, however minute, has its life-history, and it should be the aim of each student to be useful in his generation by endeavouring to furnish an accurate account of the cycle of existence of some member of the animal or the vegetable world.

Of course, before he can be expected to occupy himself with original research, he should be fairly acquainted with all that has been done before upon the subject; but above all must he be familiar with the instrument with which he works, and the methods whereby certain results may be attained.

Microscopical research does not always require the aid of expensive apparatus. It is very handy and often saves time, no doubt, to have ready all those accessories so ingeniously devised by the makers, for microscopists with long purses; but apparatus to answer the same purposes may often be made by the ingenious worker, which, though not possessing such a good appearance, serve just as well as the more expensive articles.

Many microscopists, after a few years' devotion to their favourite instrument, find themselves encumbered with a host of paraphernalia of no use to themselves or to any one else, the cost of which might have been saved by considering beforehand the capabilities of the required apparatus. As to instruments, each individual taste has to be considered, some prefer one pattern and some another; but, after all, these matters are easily arranged if the principle of construction is good.

The main office of the microscope is that of enlargement; but this amplification of the image of an object must be attained without distortion or the introduction of colours not in the original, and it is because single lenses give images blurred with spherical and chromatic aberration that double and triple combinations are used in the construction of all good microscopes. Single lenses are however very useful for general purposes: as a pocket lens, it prevents the collection of much useless material during a naturalist's rambles, and upon reaching home a further use is found in its employment as an aid in dissecting or in mounting the objects culled. The most useful magnifiers are the ordinary watchmakers' or engravers' eye-glasses in the usual horn mount, but their amplifying power should not be too great or continued use may impair the eyesight.

A combination of several single lenses, such as is shown in Fig. 1, is much used as a pocket magnifier for the field

where a high power is not required; they are cheap, but the lower powers only are generally useful. The Stanhope and Coddington lenses are also used by some collectors, though there is little doubt that they are going out of date on account of their not being so useful as newer forms of the simple microscope.

The Stanhope magnifier is a double convex lens having two unequal curvatures. In observing, the deepest curve is placed towards the eye, the object adhering to the least convex side being just in focus. The Coddington lens is generally a sphere of glass, round the periphery of which a deep groove has been cut and filled up again with black cement. This lens focusses at a short distance from the object, and is much superior to the Stanhope form. Inferior Coddingtons are now made from rejected double convex lenses which do not act as well as the form described.

FIG. 1.

Without doubt the best magnifiers for field use and such work generally are the platyscopic lenses of Mr. John Browning, which he makes of three degrees of power, amplifying 15, 20, and 30 diameters respectively. They are really achromatic triplets, are set in ebonite cells, and mounted in tortoise-shell frames. These lenses focus at about three times the distance from an object as a Coddington of the same power, and so allow of the easy examination of opaque objects. They are shown engraved full size in Fig. 2. Steinheil has produced similar lenses, which he terms "aplanatische loupen"; they are of similar construction to the above, and are made to magnify 5½, 8, 12, 16, and 24 diameters.

One of these lenses, or, preferably, two of them, carried

in the pocket when field hunting, will prove of invaluable assistance to the student. The most useful powers will be found in those amplifying 15 and 30 diameters: the former serves well for the examination of mosses, ferns, lichens, algæ, and such members of the animal world as can be

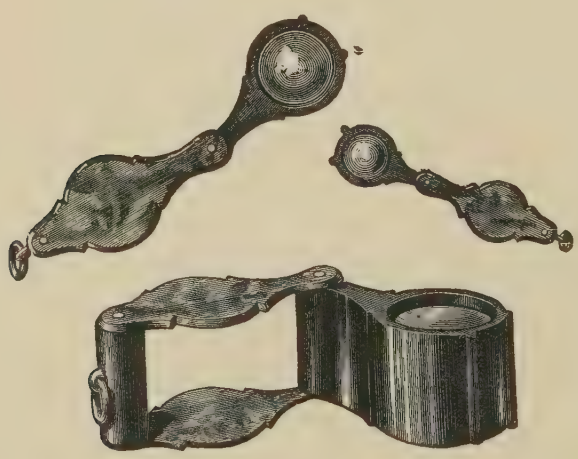


FIG. 2.

recognised by the aid of a 3-inch objective and the A eye-piece, to which combined, it is about equal in power; whilst the smaller glass, giving greater amplification, answers admirably for micro-fungi, minute algæ and lichens, and those forms of animal life for which a 1½-inch objective is desirable.

If the reader refers to any work treating upon optics, he will find that convex lenses yield images in two distinct manners—*virtual* images and *real* images. A double convex lens, when used as an ordinary magnifying glass, produces a virtual image which is erect and larger than the object, as may be seen by reference to Fig. 3.

The greater the convexity given to the surfaces of the lens, the more will it amplify, and it may also be said

that the nearer the object be placed to the principal focus of the lens (F) for parallel rays, the larger will the virtual image appear. To yield a virtual image, the object must be placed between the lens and its principal focus.

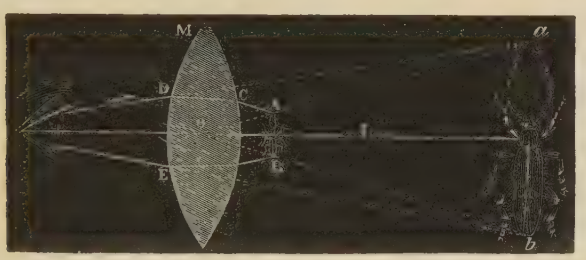


FIG. 3.

A *real* image is formed from a double convex lens, only when the object is outside, or in front of, the principal focus for parallel rays ; this image is inverted, and may be studied by receiving it upon a screen of ground glass. The action of lenses in the production of real images should be carefully considered by the student, as upon it depends chiefly

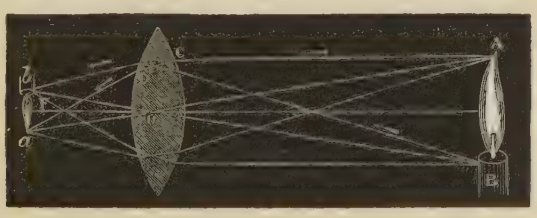


FIG. 4.

the results of his manipulation, which may be perfect or defective in proportion as he understands the theory of this simple optical instrument.

An inspection of Fig. 4 will show the two consequences of this action. If A B represents a lighted candle placed far distant from the principal focus of the lens, a small, real

but inverted image of the candle will be thrown upon the screen at *a b*, and the further A B is placed away from the principal focus for parallel rays, the smaller will be *a b*, and the nearer will it be formed to the opposite principal focus. On the other hand, if *a b* represents the lighted candle placed in front of the principal focus F but very near to it, the real image which is formed, A B, is much amplified, in proportion as *a b* is nearer to the principal focus, and the distance at which A B is formed is likewise regulated by the distance of the object from this point. The nearer *a b* is to F, the further will A B be formed away from the lens, and *vice versâ*.

The student will now be able to see the principle upon which the microscope is constructed, and, in closing this chapter, we do so by giving a table of the magnifying power of single convex lenses of varying focal lengths, for parallel rays at a distance of 12½ inches from the micrometer to the screen, which has been constructed from a paper upon the subject by Dr. Woodward in 'Silliman's Journal' for June 1872.

Focal Distance for Parallel Rays.	Amplification in Diameters.
2″	4·0
1″	10·4
½″	23·0
¼″	48·0
⅛″	98·0
1/16″	198·0

CHAPTER II.

THE MICROSCOPE STAND.

As we are supposed to be treating nearly exclusively of compound microscopes—that is to say, of instruments in which the amplification of an object is produced by means of a combination of lenses called an "objective," the image being further magnified by another set called an "ocular" or "eye-piece"—we will consider that our readers require no demonstration as to the necessity of some mechanical contrivance for holding these lenses in their correct positions.

This is the office of the stand of a compound microscope, and when we notice the progress which has been made in the details of this instrument from the time the first was made for sale in England by Mr. John Marshall, we shall find that not to opticians only, properly so called, are we indebted for improvements, but to mechanicians equally. It was but of little use attempting to perfect the optical arrangements while the mechanical contrivances were imperfect, and when this was fully appreciated, real improvements were made by mechanics and optics working hand in hand. At the same time, we must not forget the aid which has been rendered by amateurs and microscopists with unlimited means at their disposal; it is certain that many of the early and even the more recent improvements would not have been executed, except at the instance of one possessing a well-lined purse and with great interest in the science.

As previously mentioned, the chief function of the microscope stand is to receive the eye-piece and objectives;

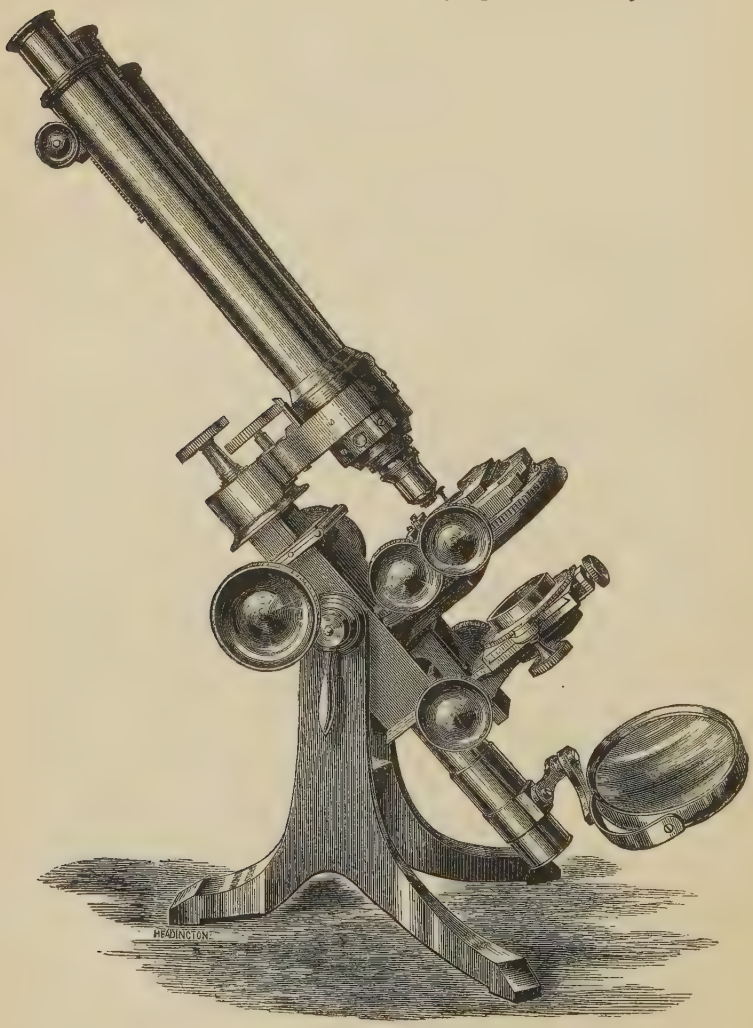


FIG. 5.

but it has to do much more than this, and upon its form and general construction, together with the character of

the workmanship, depends the excellence of the work it is able to perform.

The two principal models, upon which nearly all stands are made, have been named the "Ross model" and the "Jackson model," both of which may be studied in the productions of Messrs. Ross & Co. Fig. 5 shows the Ross model, in which the body is attached by its base to a transverse arm, which latter is borne on the summit of a racked stem, to be seen in the figure.

This is a very efficient and convenient form when the workmanship is good, and leaves nothing to be desired if the stem and transverse arm are sufficiently solid. At the same time, it is the worst model on which to make cheap instruments, as those who have used them will only be too ready to testify. A cheap stand made on the Ross model practically requires no fine adjustment, there being sufficient spring in the stem and arm to focus with, by slightly pressing upon the latter at its junction with the body.

In the Jackson model, shown in Fig. 6, the body has the rackwork attached to it, and is supported for a great part of its length on a solid limb, as shown in the figure. It is, therefore, much less liable to vibration than in the Ross model.

In a paper read before the Royal Microscopical Society in March 1870, Dr. Carpenter detailed his experience of these two forms of instruments. He sums up as follows:— "My own very decided conviction is that the adoption of the principles of the Jackson model would be decidedly advantageous alike for first-class instruments, in which the steadiness of the image when the highest powers are being employed ought to be of primary consideration; for those second-class instruments which are intended, at a less cost, to do as much of the work of the first class as they can be made to perform, portability here being of essential importance; and for those third-class instruments in which

everything has to be reduced to its simplest form, so as to permit the greatest reduction in their cost."

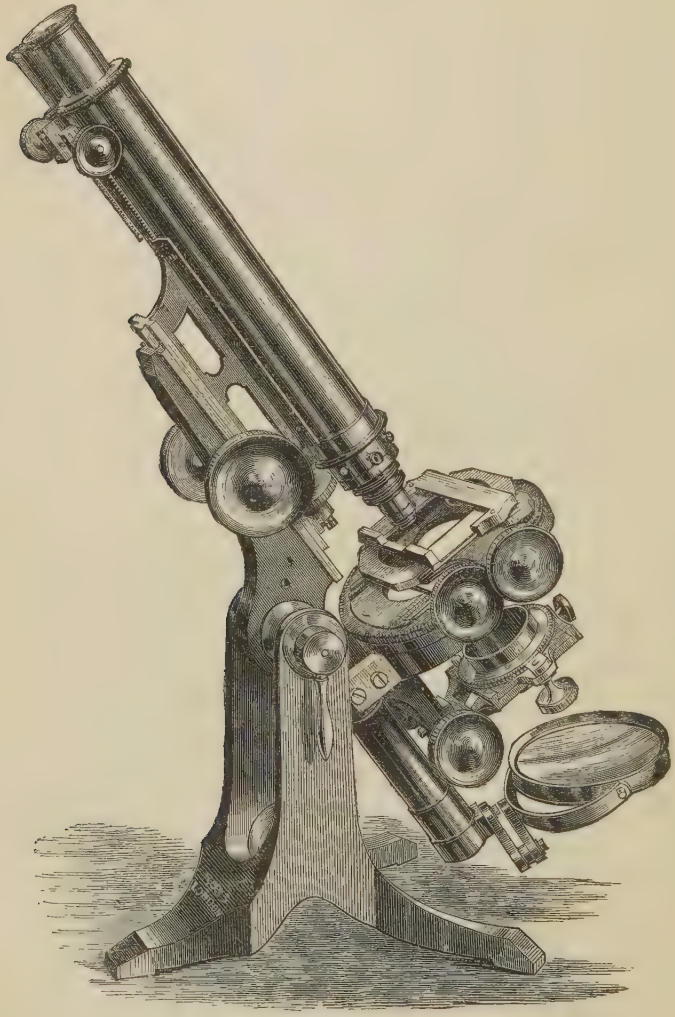


FIG. 6.

This summary of Dr. Carpenter's is so complete in its apology for separating microscope stands into three classes,

that we will not add a word more. The first-class stands are usually made from harder metal than the cheaper instruments, in which reduction of cost is an important item: this enables the former to wear better, the screws and racks allowing of a great deal of work before they begin to exhibit "backlash." The productions of Messrs. Ross & Co., Messrs. R. and J. Beck, and Messrs. Powell and Lealand deserve to be placed in the highest rank, as microscopes of the first water, and we cannot do better than describe them.

The two different patterns made by Messrs. Ross, and shown in Figs. 5 and 6, are furnished with an extremely solid foot, which is cast in one piece. The "coarse adjustment" may be easily understood from the illustration, and the "fine adjustment" is obtained by the action of a milled head upon a lever which moves the nose-piece, in either case. Both instruments are furnished with a centering and traversing substage, and also with a rotating movement which is worked by a rack and pinion. The stage itself is very complete, the object slide when laid upon it can be instantly secured in position, and the whole stage with the object *in situ* can be rotated round the optical axis as a centre. The circular motion is graduated, and thus answers many useful purposes. A rectangular motion in two directions can be imparted to the stage by means of two milled heads.

In the most recent forms of instrument the stage is made very thin (so far as is consistent with steadiness), and the central opening large, so as to admit light of great obliquity, for which purpose the mirror is placed at the end of a jointed arm, so that it may be considerably extended.

The large best microscope of Messrs. Powell and Lealand is very heavy and massive; Dr. Lionel Beale speaks of it in high terms of praise. The focussing movement is upon

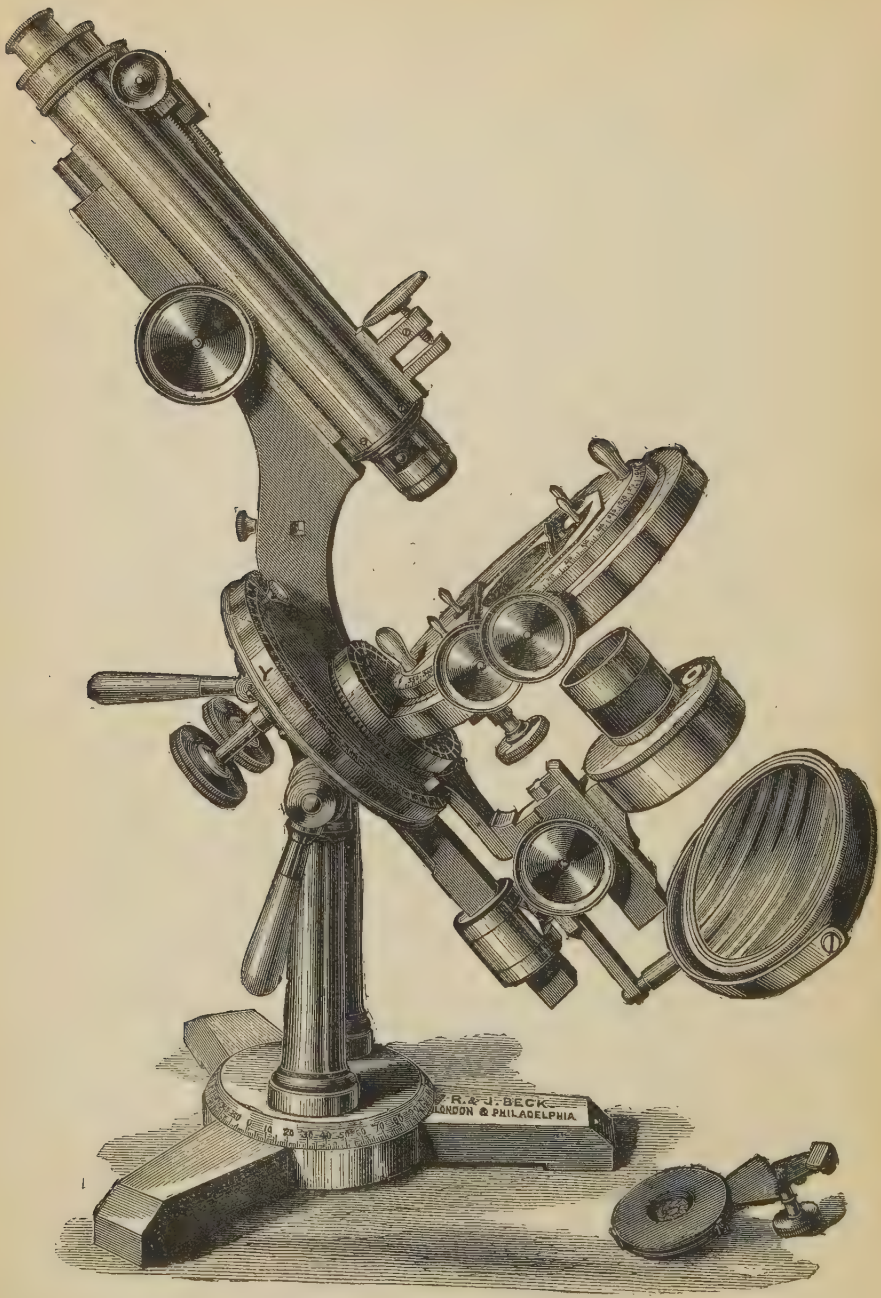


FIG. 7.

the Ross model, but the pattern itself is quite unique, and though of different construction to other forms, responds satisfactorily to all the requirements of the microscopist.

The stage has one inch of motion in two directions, by means of the milled heads, and is capable of rotation round the optical centre by means of a wheel and pinion. This is combined with a very thin stage for oblique illumination of objects, either by means of the mirror or the achromatic prism. There is also a graduated silver circle, which can be used as a goniometer or for a variety of purposes.

The substage has rotary, vertical, and rectangular motions, and both stages have for their foundation a solid brass ring, which is firmly attached to the stem. This stand is a specimen of excellent workmanship produced regardless of cost, in order to give those who require and can afford it—perfection.

With the best stand of Messrs. Beck we encounter a difficulty. It was our intention at the outset to describe those instruments furnished with swinging substages, under a special heading, but the best or "International" stand of Messrs. Beck being furnished with these additions, we feel constrained to mention it here. See Fig. 7.

All the instruments turned out from the factory of these opticians are made upon the Jackson model (by whom indeed it was first adopted) with the single exception of their "popular" microscope, which is focussed on the Ross principle. The "International" has a heavy brass tripod for its base, upon which is placed a revolving graduated fitting, from which rise two pillars, together forming the foot. Between these pillars is hung the limb which carries the body at its upper end. In its centre the compound stage is fixed, beneath which is a circular plate carrying the swinging bar and substage.

The stage itself is attached to the limb on a pivot, and

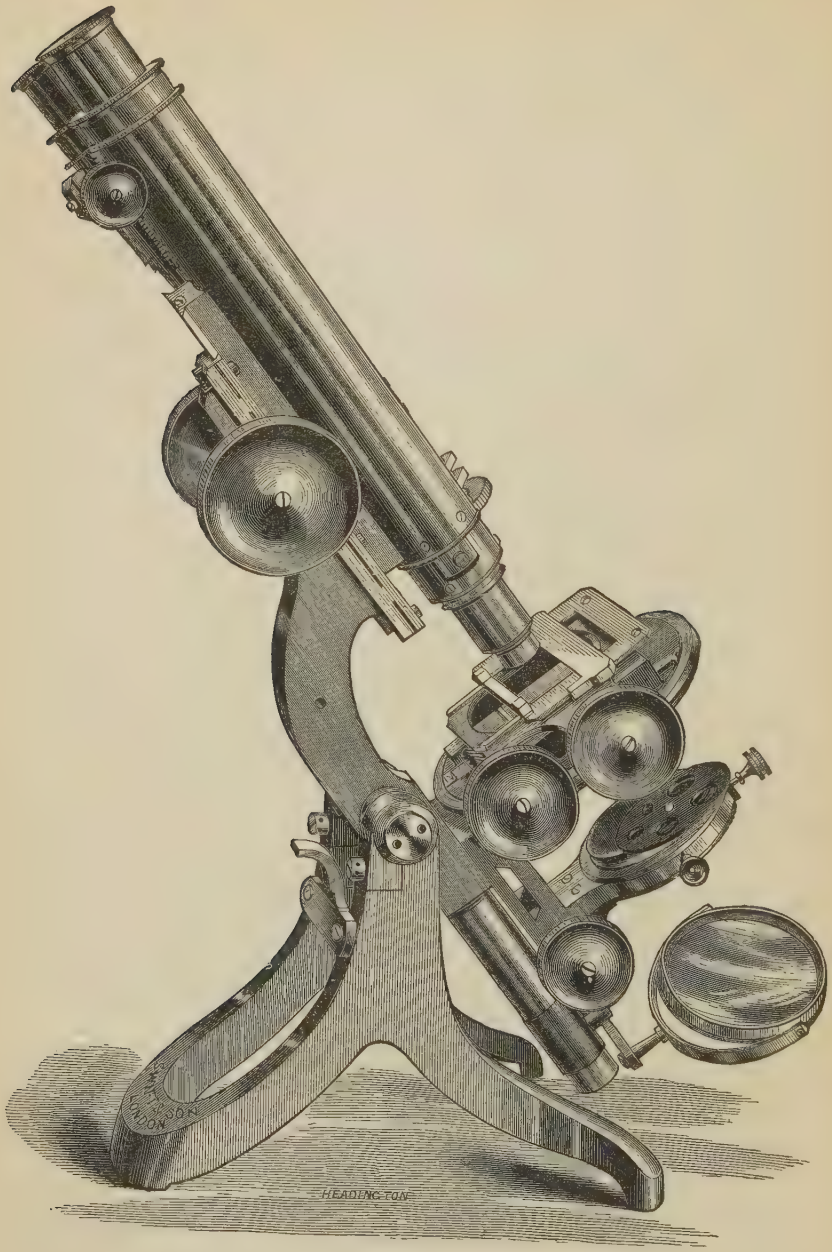


Fig. 8.

can be set to any angle of inclination, the plate being divided so that the angle may be recorded, and in this way also, light of any degree of obliquity may be used without interference from the thickness of the stage. Horizontal movements by two milled heads are capable of being imparted to the stage, which may also be revolved by means of a milled head, or when this is pulled out, by using the two ivory studs fixed to the top plate for that purpose.

For a description of the substage, we must refer our readers to a later portion of this chapter, wherein special forms of instruments are described, and in concluding a notice of these microscopes, urge every one who is wishful to possess a first-class instrument to see the work of each of these three firms before deciding. One cannot be too careful in the selection of an instrument, and we have nearly always found that if any one has rushed blindly into the purchase of a microscope on the recommendation of a friend, sooner or later he has been dissatisfied with it, and has finally purchased another of his own choice.

Other opticians produce as specialities good working stands at a medium price.

Swift's "Challenge Binocular" is shown at Fig. 8, and has been in especial favour during the past few years; it is a good working instrument, especially when combined with his universal substage condenser. The general construction of the stand will be seen on reference to the figure; it is not too tall or large, and is exceedingly well balanced.

Collins' "Harley Binocular," though formerly made upon the Ross model, is now constructed upon the Jackson model, and has this advantage, that commencing with a simple stand, a very complete instrument, such as shown by Fig. 9, can be built up from it. The stage is made to rotate and also to move by means of milled heads

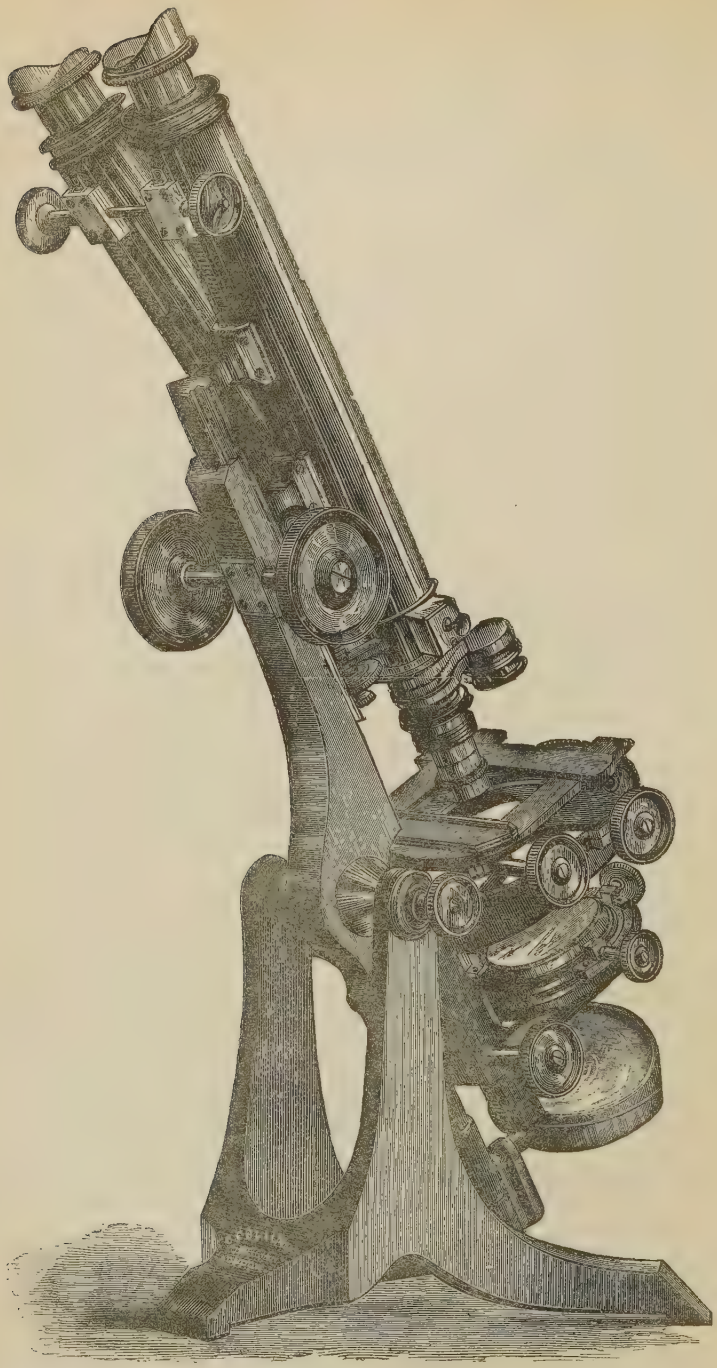


FIG. 9.

in two rectangular directions, so that the remarks made in Dr. Carpenter's 'Microscope' do not now apply to it.

Crouch's "Premier Binocular" is also one of the first-class instruments. An illustration of it is given in Fig. 10.

Messrs. Pillischer, Browning, Murray and Heath, Baker, Watson, and others make medium priced stands of considerable excellence, but seeing that all the principles of construction are exhibited in the foregoing, it would be mere repetition to give illustrations of each.

After all, a microscope stand must satisfy certain conditions, and if these be fulfilled it scarcely matters to the owner who the maker has been. It must be made of good hard brass, be furnished with a heavy foot, and well balanced, so that it may be placed in any position without overturning. All the rackwork and screws must move easily, firmly but not stiffly, and without "loss of time" or "backlash." Stops should be placed so that the body may be set either horizontally or vertically as required, and plane and concave mirrors should be always provided, preferably on a jointed arm. The lower extremity of the body should be furnished with the Society's screw, and *no microscope should be purchased which has not this thread.*

A mechanical stage is not absolutely necessary but is a great convenience, despite all opinions to the contrary, and some accessories cannot be used without it. A circular and rectangular motion should be capable of being imparted to the stage without the intervention of rackwork; but a duplicate motion by means of rack and pinion should be supplied to the better instruments. Reasons for the rotation of the stage will be found in the chapter treating on the use of the polariscope.

The medium and best stands should be furnished with an adjusting substage fitting, capable of being moved verti-

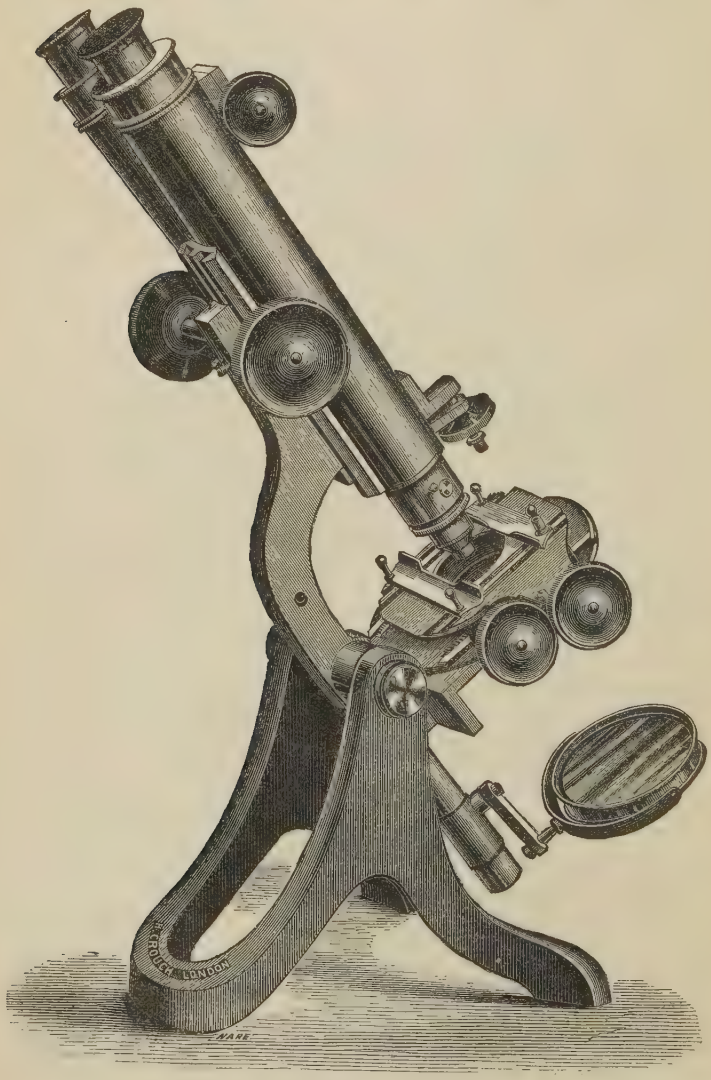


FIG. 10.

cally by means of rack and pinion; but the cheaper kinds are usually provided with a removable tube, into which the polariscope, condenser, and other accessories are made to fit. These should be capable of taking accessories made to the one and a half inch gauge, even if purchased from different makers.

A cheap but well-made microscope of special pattern has been devised by Mr. Browning, in which the entire body and stage rotates upon the same axis, perfectly concentric even with the highest powers. It has a large circular glass stage, removable stage-fitting for apparatus, and is a very good form for laboratory or general use. It is made both with monocular and binocular bodies, the former of which is shown in Fig. 11.

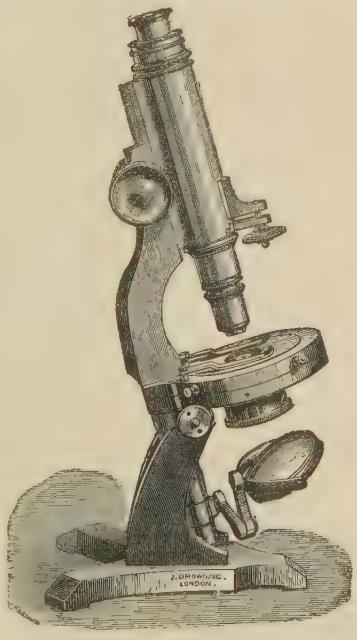


FIG. 11.

A very good form of portable microscope is made by Mr. Browning and shown in Figs. 12 and 13. The stage is fitted with rectangular and circular motions, has an adjustable substage with centering movements, and is fitted with plane and concave mirrors on jointed arm. When fitted up for use it is as steady as a stand of the ordinary make, as the author can testify, and is shown in this position in Fig. 12. By a novel arrangement the body of the microscope turns on a joint and packs away with-

out detaching any portion of the instrument, as shown in Fig. 13. The stand also folds in a novel manner and packs into a case, the outside dimensions of which are 6 × 6 × 9 inches.

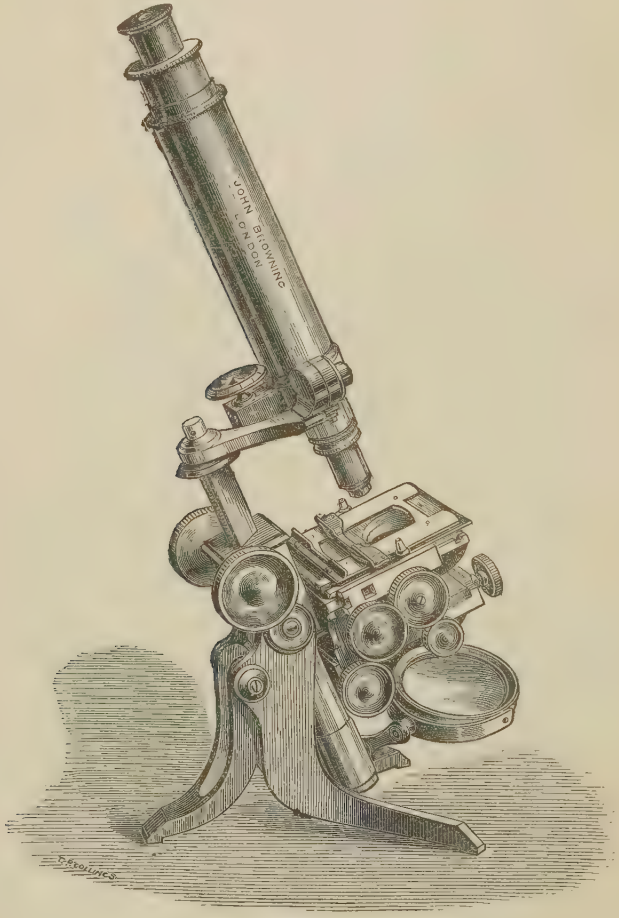


FIG. 12.

It may now be as well to notice those forms of microscope to which swinging substages have been adapted, or

those forms whereby the same effects may be obtained by other means. In 1854 Mr. Grubb, of Dublin, in a Provisional Specification for Improvements in Microscopes, stated, "My third improvement consists in the addition of a graduated sectorial arc to the microscope concentric to the plane of the object *in situ*, on which either the aforesaid prism or other suitable illuminator is made to slide, thereby producing every kind of illumination required for microscopic examination, and also the means of registering

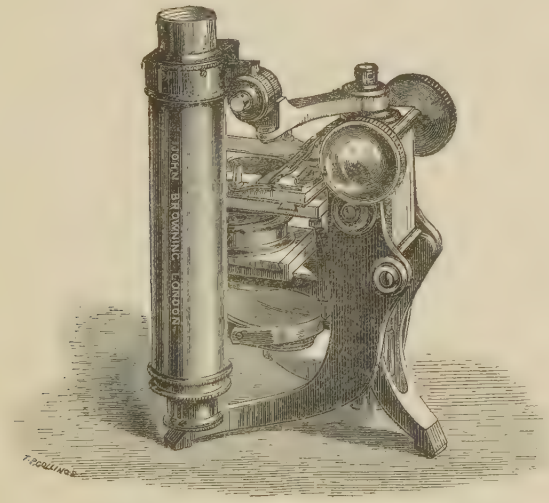


FIG. 13.

or applying any definite angle of illumination at pleasure." This seems to be the embryo of the modern swinging substage, which was not, however, developed in this country until Zentmayer, in America, produced stands with these modifications in the regular course of business.

As an instance of the pattern and the price at which these stands are sold in the United States may be mentioned the Biological stand sold by Bulloch of Chicago. The microscope stands 12½ inches high and the stage

is $3\frac{1}{2}$ inches from the table. The body is 5 inches long and the draw-tube also 5 inches in length, marked with a ring to show the standard length. The diameter of the body-tube is 1·4 inch. The stage has a revolving concentric movement, the mirror and substage can be made to swing over the tube, and there are spring stops to indicate when they are in a line with the axis of the instrument. The lower end of the main tube is fitted with the Butterfield broad-gauge screw, forming an adapter which carries the Society thread. The price of this instrument in walnut case is, with one eye-piece $40, equal to about 8*l.* 8*s.* of our money, or with $\frac{3}{4}$-inch and $\frac{1}{5}$-inch objectives 12*l.* It is shown by Fig. 14.

After the introduction of these swinging substages by Zentmayer the manufacture of them was taken up in this country by Messrs. Ross and Co., of New Bond Street, so that those who desire to be furnished with the latest improvements or modifications of the instrument can easily be supplied. The methods of construction are very similar both in the American and English instruments, though the superior workmanship of details in Fig. 15, over the Biological stand of Bulloch, is only to be expected owing to the difference in price.

This instrument is constructed on the Jackson model, and is particularly free from tremor. The coarse adjustment is effected by means of the ordinary rack and pinion, the fine movement being worked by the action of a micrometer screw acting on a lever, by means of which the body is not touched when using the fine adjustment, and as the length is not changed, the relative distance of object-glass, binocular prism, and eye-piece remains the same.

The substage bar which carries the mirror, condenser, and other substage illuminating apparatus, swings from a pivot placed behind the stage, the axis passing through the

object when *in situ*, and remains in the focus of illumination in every position of obliquity of the bar. The angle may

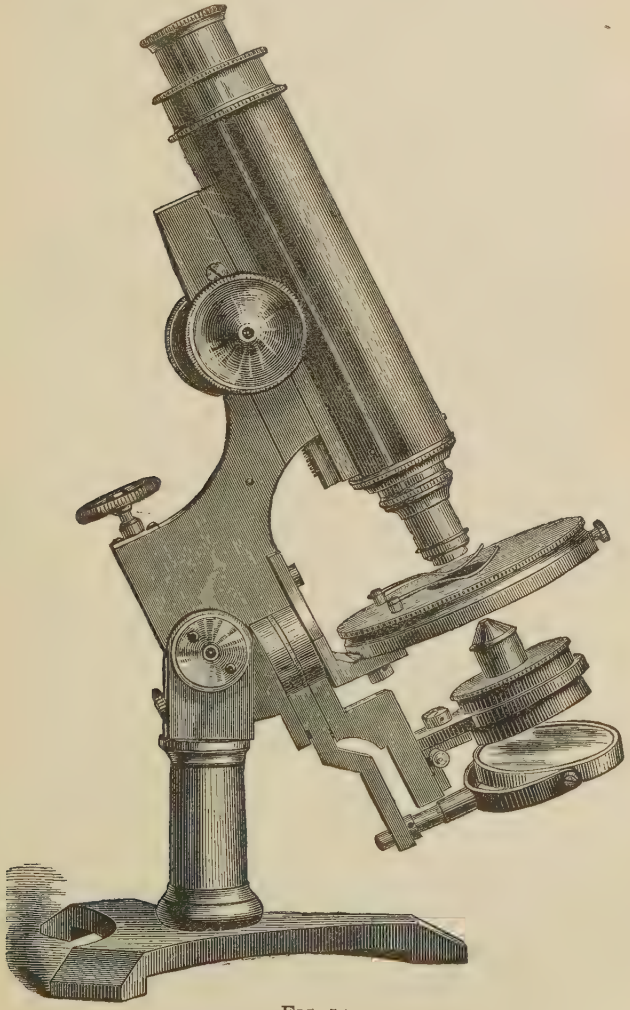


FIG. 14.

be read off from a graduated circle on a sector at the upper end of the bar.

For oblique illumination a very thin stage has been devised by Mr. Wenham ; it has only one top-plate, with rectangular and concentric rotary movements worked by two milled heads upon the same axis. This stage has two set-screws by means of which it can be easily and effectually centered. It is bevelled on the edge and graduated to serve as a goniometer. By releasing a strong clamp-nut this stage can be at once removed and any other substituted.

The swinging substage attached to the "International" microscope stand of Messrs. R. and J. Beck may be seen in Fig. 7. It is mounted perfectly true with the body, and is moved up and down in its fitting by means of milled heads. In this fitting all the varied appliances for modifying the character and direction of the light are fitted. The bar into which the substage fits is itself attached to an arc working in a circular fitting, and is capable of rotation by means of a milled head, so that it may be carried round and above the stage if necessary. The amount of angular movement may be recorded from the graduated circle. The lower triangular bar carries the mirror when the illumination is required to be concentric with the optical axis of the instrument ; when desired it can be made to slide on the substage bar, and then can be moved above or below the stage in the same manner as the substage. Messrs. Swift and Son have produced what they term a "Radial Traversing Substage Condenser," in order to produce the same results in illumination as can be obtained with swinging substages. Mention only is made here, as its construction and use will be fully explained when treating of accessories.

Messrs. Watson and Sons, of Holborn, have recently patented the arrangement, an illustration of which is shown in Fig. 16. In this the mirror is accurately centered, while

the body of the instrument, together with the stage, is made to incline in order to arrive at the same results as attained with the foregoing accessories.

It will be seen that the new stand enables the observer to

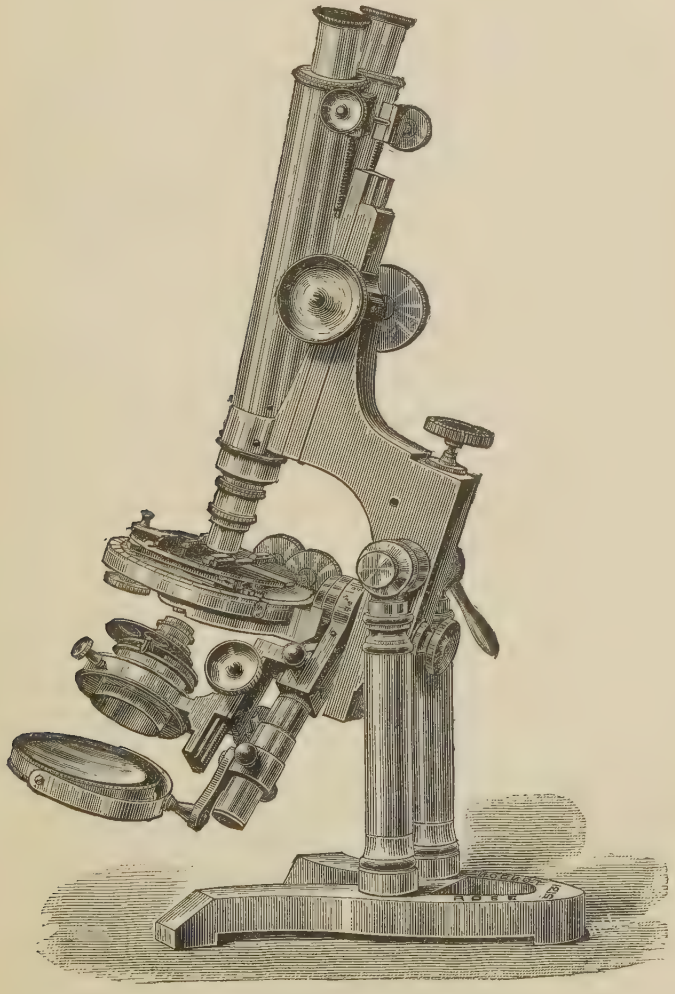


FIG. 15.

examine an object as he would if it were held in the hand, and viewed by the naked eye—that is, to turn it about in every possible way towards a ray of light proceeding in a fixed direction—and so, without once losing sight either of the light or the object, to observe its appearance when illuminated by light of every degree of obliquity.

This is the fundamental idea underlying its construction, and in this consists the great difference between it and the old forms of stand (although it has all the uses of the latter), where the object remaining fixed, the only way in which its illumination can be varied is by moving the illuminating ray—which, in the amount of the results it affords, and the amount of time it consumes, is stated by the inventors to be in every way inferior to the new one.

An inspection of the engraving (Fig. 16) will show how this idea is worked out. On the top of a strong pillar, to which it is attached by a massive cradle-joint allowing of inclination in a vertical plane, is fixed the arm carrying the body, which latter is provided with rack adjustment, and a new and improved fine adjustment, rendering unnecessary the usual often unsatisfactory loose nose-piece. The stage is so fixed with regard to the arm that the object when lying upon it is in a line with the centre of the cradle-joint, so that upon inclining the body the object moves with it, and is presented at every possible vertical angle to a ray proceeding to it from a given direction. The stage is of a new and improved construction, being exceedingly thin—in fact the thinnest mechanical stage yet devised—and is capable of giving a complete rotation of the object.

Beneath the stage swings the substage arm, concentric with the object, and carrying the usual screw centering and rack adjusting substage.

Behind the substage arm is a strong bar, provided with a dovetailed groove, into which the mirror bar slides. This

is so pivoted to the substage arm as to allow the latter to be swung aside and the mirror used alone when requisite, without the trouble of taking the substage away altogether. This is a great advantage, as it permits the substage, and any apparatus it may be carrying, to be swung into or out of position in a moment with the mirror in the position here indicated. The stand has all the uses of the old forms of microscope, and can be employed in exactly the same way, but even then its peculiar motions round the object as a centre give it very great advantages in every class of investigation. But it is when the mirror occupies the position now to be described that the peculiar properties of the new stand are brought fully into play.

The upright pillar, carrying the body and stage, is attached at its foot to a massive circular plate, carrying a graduated circle which rotates round a point exactly beneath the centre of the stage ; and moving independently and concentrically with this is another smaller circle, having a dovetailed groove ploughed across it, into which the mirror bar can be slid when withdrawn from the substage arm. A spring catch attached to the dovetailed circle falls into a notch in the mirror bar when the centre of the mirror is exactly beneath the centre of the stage. This is the most useful position for the mirror, as a ray falling from a source of light upon it may be reflected upwards perpendicularly upon the object, when the body of the microscope is vertical, then without interfering again either with the mirror or lamp, or interposing any accessory apparatus whatever, but simply by inclining the body, the light falls upon the object with a gradually increasing obliquity until, when the instrument is nearly horizontal, a perfect dark-ground illumination can be obtained even with the highest powers, while the gradual way in which the light becomes more and more oblique immediately

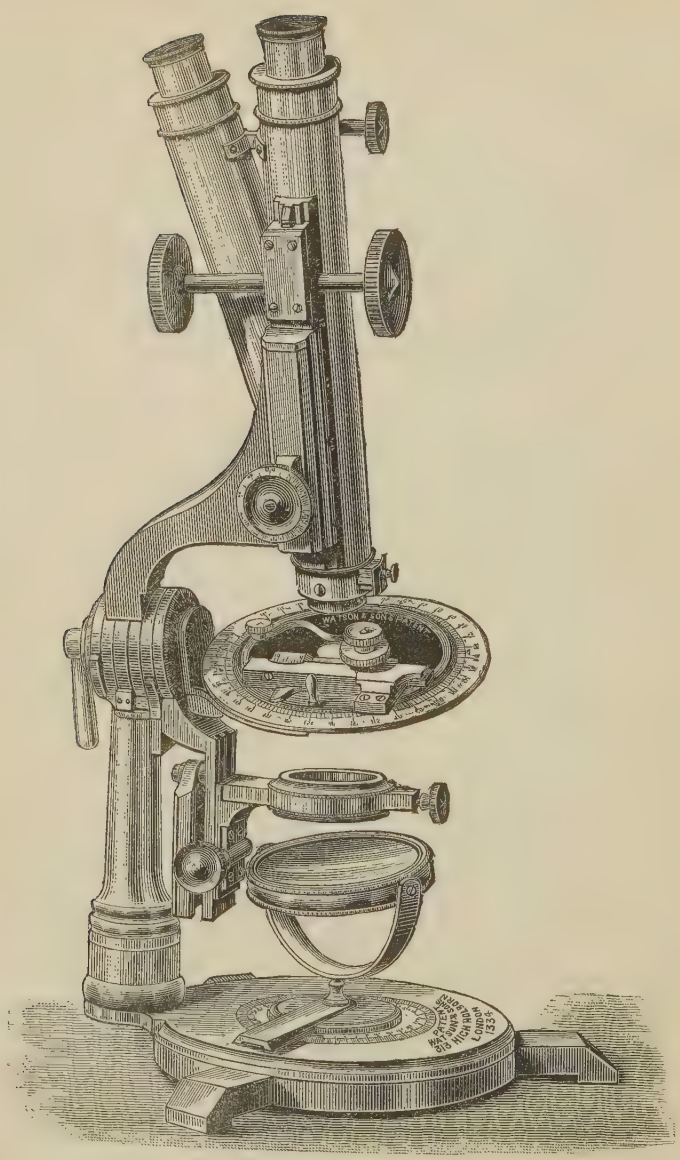


Fig. 16.

under the eye, and the capability of arresting the inclination at that point where the most suitable illumination for the object under examination is obtained, give to the observer powers he has seldom before had at his command in any form of microscope yet produced.

The horizontal rotation mentioned above allows the object to be directed to the light at every angle in azimuth—to borrow a term from the astronomer—as the cradle-joint on the top of the pillar gives every angle in altitude; the object occupying the centre of both motions, by a combination of the two it can of course be placed in every possible position. These angles are read, the latter by a graduated circle in the outer side of the cradle-joint, giving the inclination of the body to the vertical, the former by means of the graduated circle at the foot; readings of these circles being taken with the mirror placed as above described at any time by so fixing the instrument that these circles read the same. Any desired effect will be exactly reproduced, *wherever the lamp may be placed*—a point of the greatest importance to workers with high powers.

There is a third divided circle on the substage axis, giving the inclination of the substage to the axis of the body. A strong clamp on the outer side of the cradle-joint holds the body firmly at any inclination, and a graduation on the slide of the coarse adjustment enables the working distance of the objectives to be measured and compared.

Time alone can show whether these present apparent advantages are lasting, or whether the instrument will wear as well as those of the old form of construction.

The foregoing illustrations of microscope stands are seen to be both monocular and binocular—that is to say, used with one or two eye-pieces. A binocular should always be purchased when the student's means are sufficient, as most objects can be studied with much more ease and comfort

than with a monocular microscope. Binocular instruments are made in such a manner that the rays proceeding from an object lying upon the stage are split into two parts, each portion passing separately through a tube to the eye-pieces, as shown in Fig. 17.

The form of prism used for dividing the rays (we may say universally) is that devised by Mr. Wenham many years ago. It is mounted in a small brass box, sliding into the lower end of the microscope body immediately over the objective, as shown in Fig. 17. A section of this prism is shown in Fig. 18, the dotted line indicating the direction of one portion of the divided ray.

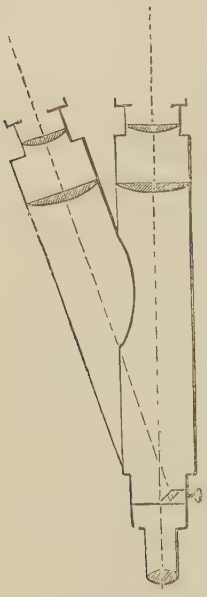


FIG. 17.

It should not be forgotten that a binocular of this form is

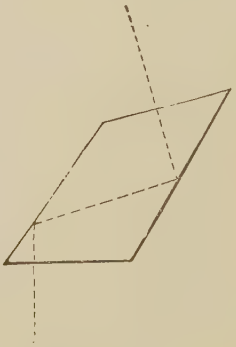


FIG. 18.

not well suited for use with objectives of a higher numerical aperture than 0·34 or 40° air angle; a certain amount of distortion occurs with higher apertures, which may be readily perceived on the examination of spherical pollen-grains and such like objects. Dr. Carpenter has pointed out this exaggerated relief years ago, yet there are some folks now

who delight in showing this imperfection as one of the beauties of the binocular microscope.

It will thus be seen that objectives of higher amplification than the half-inch cannot be satisfactorily used with the binocular, and even the half-inch requires making of specially small aperture to adapt it to this use. The prism is always made removable, so that at any time the microscope can be used as a monocular by simply withdrawing it.

In order to enable observers to use binocular vision with high powers, Messrs. Powell and Lealand have devised a system of prisms which may be used with objectives as high as the $\frac{1}{50}$. The images produced are not stereoscopic, but exactly similar. Mr. Wenham has also devised prisms for using high powers binocularly, which appear to act in a very successful manner.

Tolles, of Canastota, U.S., makes a binocular eye-piece for the ordinary single body, which gives a large and well-illuminated field with low and medium powers. Professor H. L. Smith remarks that he has used the $\frac{1}{12}$ and $\frac{1}{16}$ objectives with it.

Another binocular microscope was devised some years ago by Mr. J. W. Stephenson, which has since been successfully made by Mr. Browning. Within the last few years the cost of this instrument has been considerably reduced, Messrs. Swift and Son and Mr. Baker having each constructed a Stephenson binocular for dissecting purposes at the moderate price of *7l.*

The Stephenson binocular, as made by Mr. Browning, is shown at Fig. 19, in which the change from binocular to monocular or *vice versâ* can be effected without unscrewing any part or interfering with the object under examination.

In this form of binocular the rays are divided by two

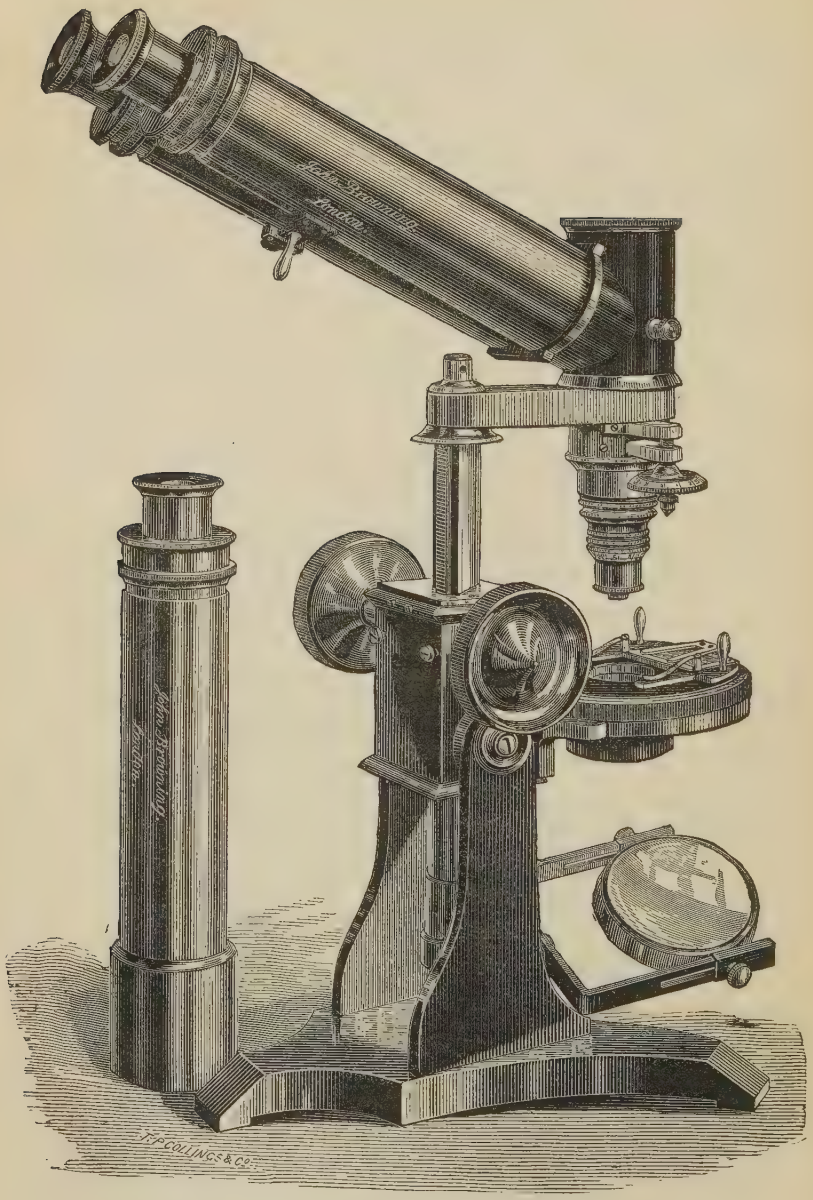


FIG. 19.

prisms A A, Fig. 20, which, passing upwards, are reflected towards the eye-pieces by the triangular prism B, Fig. 21. The instrument is erecting—that is to say, objects are presented to the eye in a normal manner, and not inverted as in the ordinary form of instrument, which adapts it specially for use in dissecting. The bodies of the microscope are made to rotate, carrying the prisms with them, so that two observers may work with the instrument,

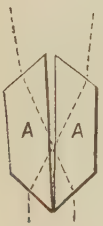


FIG. 20.

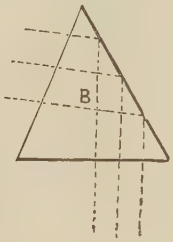


FIG. 21.

observe the same object, and compare notes without altering the conditions under which the object is being examined.

Amongst the foregoing medium and best stands, the reader must be fastidious indeed if he is not able to find one to suit his choice. Price might be a deterrent, and therefore some illustrations are given of several microscopes which, though cheap, are good working instruments.

Fig. 22 is the Economic monocular microscope of Messrs. R. and J. Beck, which with coarse and fine adjustment, 1-inch and $\frac{1}{4}$-inch objectives, two eye-pieces, concave mirror, side condensing lens, diaphragm, forceps, pliers, and glass slip with ledge, in mahogany case, costs 6*l.* 12*s.* 6*d.* A small cheap stand such as this is nearly always required by the practical microscopist, even if he possesses a first-rate instrument; for travelling, Society

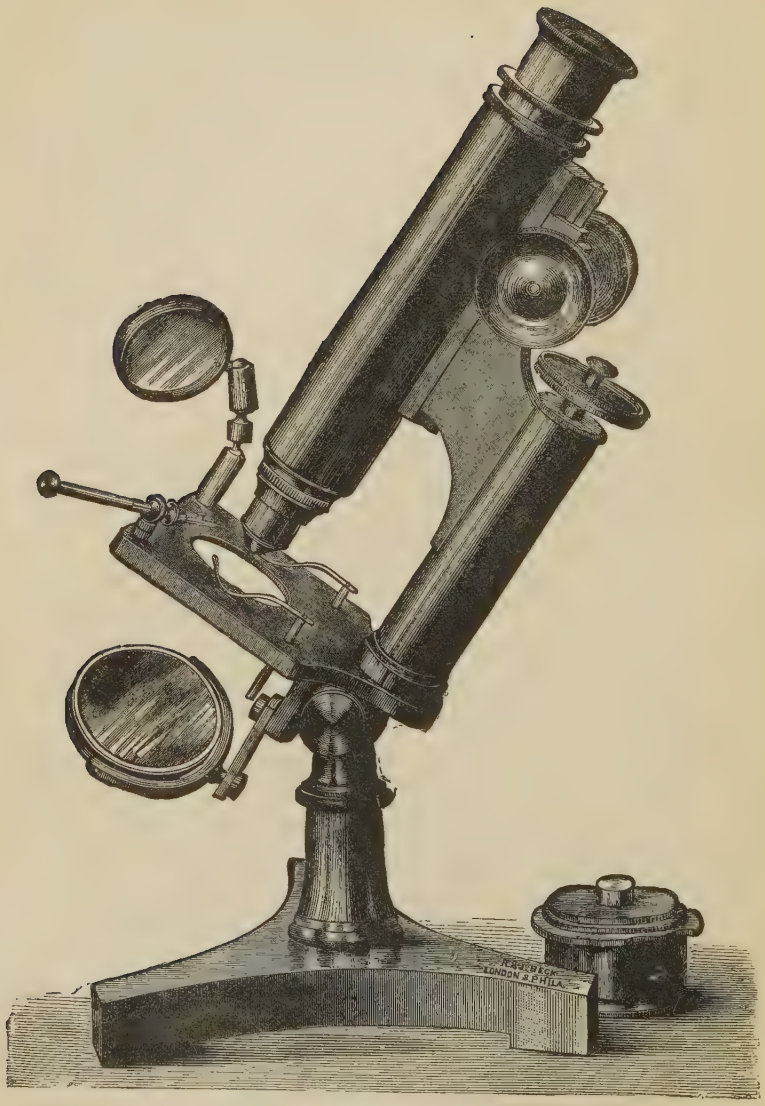


FIG. 22.

demonstrations, dissecting, and many other purposes a small microscope is exceedingly useful.

Another cheap instrument, which is furnished by several makers, is shown in Fig. 23; it has a sliding body for coarse adjustment, a fine adjustment, draw-tube, wheel of diaphragms, tube fitting for substage apparatus, plane and concave mirrors, one eye-piece, 1-inch and $\frac{1}{4}$-inch objectives, and stand condenser, in mahogany case, for which is charged the moderate price of 5*l.* 12*s.* 6*d.*, to which can be added afterwards a polariscope for 25*s.*, a camera lucida 6*s.*, and a stage micrometer for 5*s.* 6*d.*

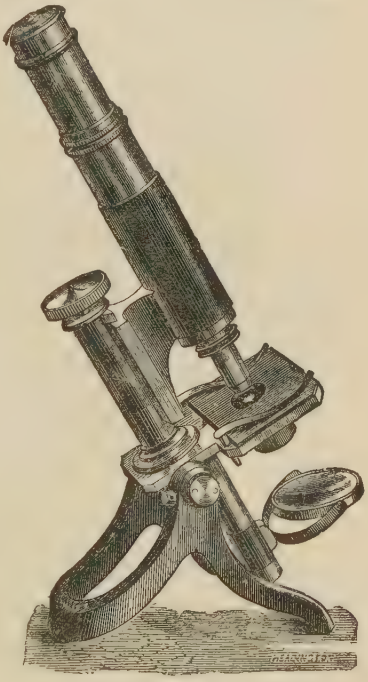


FIG. 23.

Equipped with one of these instruments, which are remarkably steady, being hung on the Jackson model, the student is ready for an immense amount of work, and it is certainly much better to invest in an instrument such as this than to waste money on a cheaper instrument which has not the Society's screw. In the purchase of a low-price stand, we would give the student the following advice:—Never purchase a cheap instrument made upon the Ross model; never purchase one which will not take objectives with the Society's thread without an adapter; never purchase an instrument without a fine adjustment,

without it be for petrological studies merely; and above all do not purchase the extremely trashy foreign separating objectives. If you want foreign glasses, there are good ones to be bought—buy them.

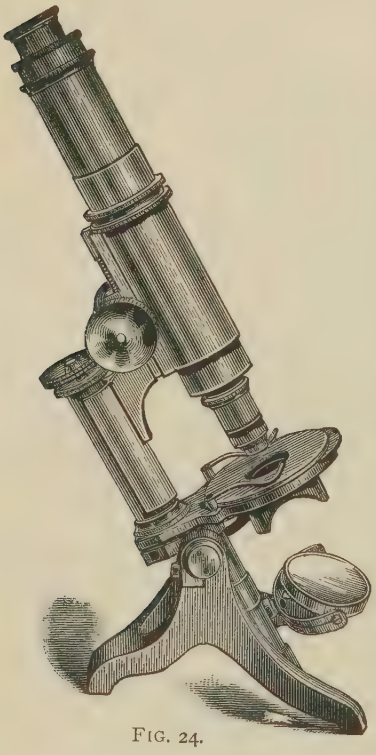


Fig. 24.

After all, there are a few objections to the form of instrument shown at Fig. 23. The body tube is narrow and the field lens of the eye-piece small in proportion, so that the field is limited in size; then, again, the absence of a coarse adjustment may for some purposes be found inconvenient, while the fitting below the stage should be made removable. On the other hand, the short tube gives a larger field when used for photo-micrography, and is also more convenient as a dissecting microscope, lengthening as it does the anterior conjugate focus and giving more room for the needles.

Recognising these and several other advantages, the author had one specially constructed, as shown in Fig. 24. The draw tube is wide enough to take the full-sized eye-pieces, and when it is fully extended the whole forms a body of the ordinary length. The stage, together with the body, may be rotated round the optical axis in the same

manner as has been already shown in Fig. 11. There are coarse and fine adjustments, a diaphragm in the thickness of the stage, plane and concave mirrors, the whole costing the author the moderate sum of three guineas and a half.

With this form one is not bound to bad or medium eye-pieces and objectives; they can be bought separately and selected. We should like to see some enterprising manufacturer making the instrument as shown in Fig. 24, and selling it for six guineas with an A eye-piece and a good inch objective, or without eye-piece and objective for 3*l.* 10*s.* The details of this instrument are as follow:—When standing vertically and closed down, the top of the eye-piece is 11 inches from the table, the collar in which the body slides being 3 inches in length, and lined with velvet. The body is 5 inches in length, and the draw-tube $4\frac{1}{2}$ inches, with an outside diameter of 1·3 inches; the stage is 3·7 inches from the table, and has a diameter of 3 inches; the mirrors are 2·2 inches in diameter, and the fine adjustment raises or depresses the entire body $\frac{1}{100}$ of an inch for each complete revolution. The stage is less than $\frac{1}{4}$ of an inch in thickness. The author has found this a very convenient instrument for photomicrography and for general microscopical work. The substage fitting will take all accessories made to the usual $1\frac{1}{2}$-inch gauge.

It is greatly to be deplored that there exists no universal gauge for eye-pieces and substage fittings. Is it too much to ask the Council of the Royal Microscopical Society to take the matter in hand?

CHAPTER III.

EYE-PIECES AND OBJECTIVES.

Eye-pieces or Oculars.—When the student purchases a microscope stand, he will generally find it supplied with the lowest power Huyghenian or negative eye-piece, usually designated by the letter A. At the same time, it may be stated that others, possessing greater degrees of amplification are often substituted or added at the wish of the purchaser; and it should be remembered, in the selection of a microscope stand, that the eye-pieces of one maker will not always fit the tubes made by another. It is a thousand pities opticians have not yet learned that their time may be more profitably occupied than by making adapters for each other's instruments.

The Huyghenian eye-pieces or "oculars" of low power are generally styled "shallow," to distinguish them from those which give greater amplification, which are called "deep"—the terms deep and shallow being applied to the degree of curvature possessed by the lenses employed in their construction, and not to the distance between them, as some writers have stated.

A full-size section of the Huyghenian A eye-piece is shown in Fig. 25, so that the student may understand the details of its construction.

It consists of two plano-convex lenses, placed at a distance from each other equal to half the sum of their focal lengths, the best proportion of relative radii being 1 to 3.

The lower lens is called the field-glass, and the upper one the eye-glass, while a circular stop or diaphragm is placed nearly midway between the two.

The practical optician Gundlach has stated that the correction afforded by the Huyghenian eye-piece is not a complete one; for at the point where the spherical aberration is entirely corrected, the chromatic has not completely disappeared. This even at the most favourable interval between the two lenses.

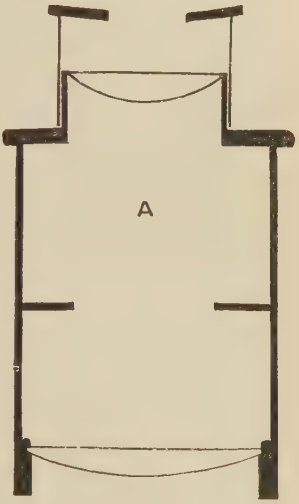


FIG. 25.

This eye-piece was first employed by Huyghens for his telescopes, in order to diminish spherical aberration and to increase the size of the field. An elaborate dissertation upon it has been published by Mr. Varley in the 'Transactions of the Society of Arts,' vol. li., to which the student is referred. It is often called the negative eye-piece, on account of its correcting the positive aberrations of the objective.

In Fig. 26 is shown a section of a deep eye-piece—the Huyghenian C—in which it will be seen that the lenses possess deeper curvature than in the A, while the diaphragm is more contracted, and the aperture in the cap covering the eye-glass is very small indeed; the C eye-piece gives about double the amplification of the A.

There is another kind of ocular in occasional use, called Ramsden's positive eye-piece; it is formed of two plano-convex lenses, but the curvature of the field-glass is turned towards the eye instead of towards the object, as in the

Huyghenian. In this eye-piece the focus is obtained *in front* of the field-glass, while in the Huyghenian ocular the image is formed *at the diaphragm*, about midway between the field-lens and eye-glass.

The Ramsden eye-piece was much in use at one time for purposes of micrometry, as it gave an excellent view of the micrometer, free from distortion even to the edges of the field, though the image was slightly coloured. It is still used by Messrs. Ross and Co. for their eye-piece micrometers.

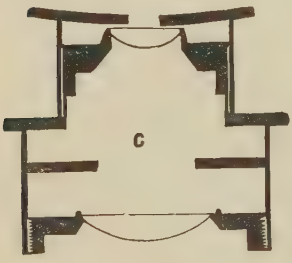


FIG. 26.

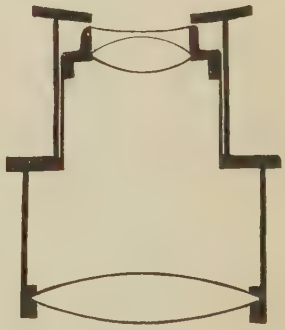


FIG. 27.

Kellner's orthoscopic eye-piece is much employed where a large and flat field is required for use with low powers. A section is shown in Fig. 27, from which the student may gather that the field-glass is doubly convex, and the eye-glass a slightly under-corrected achromatic combination, while the diaphragm is dispensed with altogether.

In the ordinary or Huyghenian ocular, English opticians designate their power by means of letters, A, B, C, D, E, and F, while some few call their productions by the numbers 1, 2, 3, 4, and 5 ; they seem fairly agreed as to what should be the relative degrees of amplification of the A and B eye-pieces, and some with the A, B, and C ; but with higher powers there seems no uniformity, as the following

table will show. The numbers have been calculated from the catalogues of the different makers:—

Objectives.	A	B	C	D	E	F
Ross	1·0	1·6	2·5	4·0	5·0	8·0
Powell and Lealand ..	1·0[1]	1·5[2]	2·0[3]	4·0[4]	6·0[5]	
Beck	1·0	1·6	2·6	3·2	5·2	
Swift	1·0	1·4	2·0	2·8	4·3	5·6
Browning	1·0	1·3	2·0	3·7		
Watson	1·0	1·4	2·0	2·5	3·4	4·3
Crouch	1·0[1]	2·0[2]	3·3[3]			
Collins	1·0	1·6	2·5	4·0		
Baker	1·0	1·6	3·0	4·5		
Pillischer	1·0[1]	1·6[2]	3·0[3]	3·5[4]		
Parkes	1·0	1·5	2·2			
Zeiss	1·0[1]	1·3[2]	1·8[3]	2·6[4]	3·5[5]	
Hartnack	1·0[1]	1·2[2]	1·5[3]	2·3[4]	3·0[5]	3·5[6]

Our American brethren treat their oculars in a more rational manner, they style them as "2-inch," "1-inch," or otherwise, as the case may be, according to the degree of amplification they yield when compared with single lenses; thus a "2-inch ocular" would amplify the same as a single lens of 2-inch focus, and so on in like proportion.

Now reference to Chapter I. will show us that a single lens of 2-inch focus (equal to the English A eye-piece) magnifies about five diameters at a distance of 12½ inches from the micrometer to the screen, and the 1-inch, 10 diameters (equal to the C eye-piece), so that it is well the student should as early as possible grasp the fact that the amplification of an object is arrived at by two stages, the objective producing an enlarged image of the object, which the ocular magnifies still further. Roughly it may be stated that if the inch objective be used with the 1-inch ocular an amplification of 100 diameters is arrived at, the objective magnifying the object 10 diameters, the image of which is further magnified 10 diameters by the ocular; and further, if the same objective

be used with the 2-inch ocular (the A eye-piece), the former will produce an enlarged image of 10 diameters, which the ocular will again magnify five times, producing a total amplification of fifty.

This is not mathematically exact without every disturbing element be taken into consideration, but is quite near enough to illustrate the case in practice. Fifty diameters is the recognised amplification for the 1-inch objective combined with the A ocular at a distance of 10 inches, and as the enlargement of the object takes place in two distinct stages, it will be seen that the optician is able to vary the powers of both ocular and objective and still obtain the standard result. An inch (so-called) objective magnifying 8·3 diameters when used with an A eye-piece magnifying the image six diameters, will give a normal result, as will an objective of the same designation magnifying 12·5 diameters with an A ocular magnifying four.

Similar cases to both of these have recently fallen under the notice of the author, and it is on account of like departures, that abnormal amplifications are obtained when the objectives of one maker are used with the oculars of another.

It often happens with new oculars that particles of brass get detached, and fall upon the inner surface of the lenses, which must be removed by unscrewing, and then *carefully* wiping with a *very soft* wash-leather. Dust specks and bubbles may be easily detected by deflecting a dull light through the body of the instrument, when, by observation during the rotation of the eye-piece, they show very plainly. A good eye-piece should be perfectly transparent, and free from striæ and markings and spots of any kind. The marginal circle of the field of vision should be sharp, clear, and intensely black. If these conditions are not fulfilled, the eye-piece cannot be considered as perfect, or fit for general use.

Before proceeding to treat of objectives, the student may be advised to commence with the 2-inch ocular (an A). If he wishes for more than one power, the 1-inch ocular (C) is recommended, it being double the power of the A. An orthoscopic C would perhaps be useful.

Object-glasses or Objectives.—The history of the achromatic objective is a curious one—interesting certainly, but it should teach us the serious lesson not to be dogmatic in our assertions. Biot and Wollaston, the latter especially, were wedded to doublets, and they both predicted, on the faith of certain experiments which were then unsuccessful, that the compound microscope would never excel the simple. How far this prediction has been verified most of our readers will know; but it is certain that Wollaston never thought that within fifty years of his prediction the doublet would be a thing of the past, rarely heard of and never seen.

Light seems to have dawned upon objective construction through the elder Dollond, who employed two different kinds of glass in the construction of his telescopes. Recognising this principle, several foreign opticians made partly corrected glasses as early as 1824, and at the same time Tulley of London produced the first achromatic objective made in England: it was composed of three lenses and possessed an air angle of 18°, which he soon after increased to 38° by placing another corrected combination in front of it.

In the year 1829, Mr. Joseph Jackson Lister, in his celebrated paper published in the 'Philosophical Transactions of the Royal Society,' pointed out how many of the difficulties could be overcome, and exhibited an objective of 50° air angle which gave a large field and a correct image. This advance was so great that it astonished Dr. Goring, who wrote, in his 'Exordium to Microscopic

Illustrations,' that "microscopes are now placed completely on a level with telescopes and, like them, must remain stationary in their construction."

Improvements, however, continued to be effected; Mr. Thomas Ross, upon increasing the air angle, discovered that different thicknesses of covering glass disturbed the corrections for spherical and chromatic aberrations no matter how carefully made, and in 1837 he presented a paper to the Society of Arts upon the subject. In this paper he stated having made an improved combination, the focal length being one-eighth of an inch, with an air angle of 60°. After this he announced obtaining an air angle of 135°, and falling into a similar error of dogmatism as Goring, Wollaston, and Biot, stated that "135° is the largest angular pencil that can be passed through a microscope object-glass."

In 1851, Chas. A. Spencer, of Canastota, N.Y., produced objectives of 146° air angle, and in 1857 he constructed a one-twelfth with an angle of 178°. Since this, Mr. Tolles, of Boston, has made lenses claiming to be infinitely near 180°, and this angle in air has been approached by several of the best English makers.

But these are not the whole of the improvements which have been effected; air lenses or dry objectives have been supplemented by water-immersion powers, and finally we have the homogeneous-immersion system, in which the transmitted ray pursues a rectilinear course from the under side of the object slide until it leaves the posterior surface of the front lens. These immersion lenses will be fully described later on; all we wish to state here in reference to them may be said in the words of Prof. Abbe: "A wide angle immersion glass may therefore utilise rays from an object in a denser medium which are entirely lost for the image—which in fact do not exist—when the same object is in air, or is

observed through a film of air;" and again, "Consequently we have a *loss* of aperture when an air angle of 180° is substituted for a balsam-angle of 100°," for "an immersion objective of balsam angle exceeding twice the critical angle (41°) has a *greater aperture* than any dry lens can ever have." It was upon this subject that Mr. Wenham fell into error and so may be classed with Ross, Goring, Wollaston, and Biot. He denied that Tolles had produced, or could ever produce an objective of greater balsam angle than was equivalent to infinitely near 180° measured in air; and those who remember the correspondence on this subject which appeared in the pages of the 'Monthly Microscopical Journal,' will now smile when they see the announcements in the 'Journal of the Royal Microscopical Society' of the productions of Messrs. Powell and Lealand of 150° balsam angle. That memorable correspondence should teach us to remember how easy it is to fall into error, and also that it is quite easy to persuade ourselves we are well acquainted with our subject, while at the same time we may totally misunderstand it.

Objectives — which, by-the-by, are sometimes called "powers"—being made from glass of varying density and also of varying refractive indices, it follows that they must differ also in construction in some degree. The various lenses of which each combination is constructed are ground to a series of curves, suitable to the glass employed, and the combinations are placed at different distances apart, so that we can only give a rough outline of their general construction.

As a rule, objective mounts are turned out much too long. There is no apparent reason why the brasswork (of some opticians especially) should not be considerably reduced. When the posterior lens is too far away from the Wenham prism, in a binocular instrument, it is

extremely difficult to procure an equal illumination of the whole field—in fact, the general performance of the instrument is seriously interfered with, and therefore, for use with the binocular, short mounts should be preferred and the longer ones rejected.

Mr. Swift has recently issued a series of objectives in short mounts, especially for use with the Wenham binocular, each of which allows of perfect illumination, without the aid of an achromatic condenser. They range from 1 inch upwards.

Objectives are generally spoken of in terms of the amplification which they yield, the standard of comparison being the magnification given by a single lens of the nominal focus. The student must not, therefore, imagine that an objective stated to be 1-inch, ½-inch, or so on, will focus at these distances from the object. Opticians have never used the term in that sense, though a few writers in public journals seem to have understood the nomenclature in that light.

The degree of amplification, taken with the aperture, is the surest guide to the focal distance; for as the aperture increases, the less will the working distance be, a point to be considered in the selection of an objective for students' use.

The lower powers are often made to separate. An objective giving the amplification of a 2-inch when complete, is converted into a 4-inch by removing the anterior combination. Separating 2-inch and 1-inch, 1-inch and ½-inch, ½-inch and ¼-inch, are also made, with others; but the student is advised not to invest much in these separating powers.

The best low-power objectives for general work will be found to possess the following air angles:— 9° for the 4-inch, 15° for the 2-inch, while the 1-inch seems to perform most satisfactorily at 30°.

No objective with an air angle of more than 40° should be used with the Wenham binocular. Dr. Carpenter pointed out long ago the exaggerated effect of projection produced when pollen-grains of the *Malvaceæ* and other similar objects are examined binocularly with high-angle objectives; perfectly spherical objects, instead of resembling a hemisphere, appearing like the small end of an egg.

Powers yielding amplifications ranging between 50 and 200 diameters may be called medium, and are represented in our list by the ½-inch and ¼-inch objectives. The former is a very handy glass, though it does not seem to be an easy one to construct, judging from many the author has seen. Makers of cheap but really good 1-inch and ¼-inch powers seem to fail sometimes in producing cheap and perfect ½-inch objectives.

A good working ½-inch may have an aperture of 60°, though it is made of 35° by Mr. Browning, and 40° by Messrs. Powell and Lealand and Mr. Collins, for special use with the binocular.

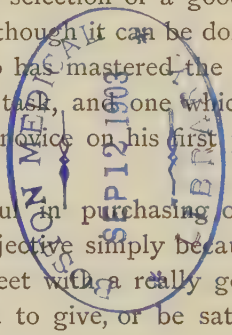
The medium and higher powers should be chosen with conical fronts, as shown in Fig. 30, for with them it is more easy to illuminate opaque objects satisfactorily. The illustration is that of Mr. Swift's short mounted ¼-inch objective, with collar adjustment.

Zentmayer, Wray, and Zeiss have produced low power objectives in which the two combinations composing them are separated or brought nearer to each other by means of a screw collar, the lens being nominally a 4-inch, a 2-inch, or any intermediate power at will. This glass defines well, and, moreover, possesses a flat field; but, according to measurement, the amplification it yields more nearly approaches a 5-inch and 3-inch than an English 4-inch and 2-inch.

For several years cheap foreign lenses were in great

request, and much work has been done with them. This has induced several English makers to offer a series of low-angle objectives at a cheap rate, in addition to the really good glasses which the same opticians turn out. Browning, Wray, Watson, Swift, Dancer, Collins, and Parkes make very excellent and cheap lenses for histological work.

It sometimes happens that very good powers may be met with second-hand, or from the workshops of neglected makers; but the selection of a good glass from several of average quality, though it can be done by an optician, or a microscopist who has mastered the elements of science, is not a very easy task, and one which certainly cannot be performed by a novice on his first purchase of an instrument.



Be very careful in purchasing objectives. Don't buy trash, or any objective simply because it is cheap. Wait until you can meet with a really good glass at the price you are disposed to give, or be satisfied with the powers you already possess.

For botanical and general work, especially if the student's means are limited, the inch and $\frac{1}{4}$-inch objectives will be found sufficient; the 2-inch, and afterwards the $\frac{1}{2}$-inch, may be added, if required; following, if necessary, with the $\frac{1}{8}$-inch and $\frac{1}{16}$-inch. A 4-inch power is used by some for the examination of wood sections, whole insects, etc. A cabinet will generally contain the following objectives by the time the owner considers it furnished:—4, 2, 1, $\frac{1}{2}$, $\frac{1}{4}$, $\frac{1}{8}$, $\frac{1}{16}$, which yield degrees of amplification varying from 12 to 800 diameters with the A eye-piece, and 24 to 1600 with the C.

The low powers are constructed in several ways, according to the aperture desired. Those of greater amplification than the 1-inch are made either as triplets or of two pairs of lenses placed at certain distances apart, as all

the corrections required are easily made on this system. The triplet, used only for low angles, is the least to be commended, and should not be used with deep eye-pieces. The 1-inch objective may be a triplet, as shown in Fig. 28, or a double combination, as in Fig. 29.

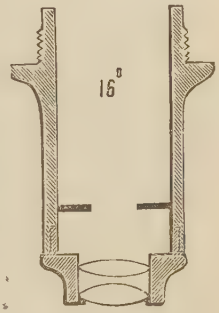


FIG. 28.

In the latter the front is a plano-convex of crown, with a meniscus of flint, being separated by a considerable interval from the posterior combination, which is composed of two double convex crown lenses, holding between them a double concave of flint.

It will be seen from Figs. 28 and 29 that there is more work in the construction of the higher angle. The cost is consequently greater; but when we remember that the 1-inch of 25° admits more light than the one of 16°, that it defines better, resolves better, and proves to be a much superior working glass in every respect, the extra money will not perhaps be grudged for it.

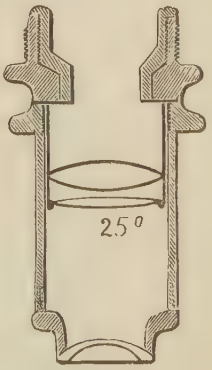


FIG. 29.

Half-inch objectives are made on two systems: the low angles for binocular use of a thick solid front, at the back of which are two pairs of partly corrected lenses, the aberrations being finally corrected by the thickness of the front. The higher angles are constructed of three pairs of lenses, the posterior combination being of considerable width. They are made of as high an aperture as 0·66 or 82° air angle, while the $\frac{4}{10}$ths of 100° air angle, or numerical aperture 0·76, is far from uncommon.

The $\frac{1}{4}$-inch objective, yielding a power of 200 diameters with the A eye-piece, is a most useful glass to the student, when possessing an air angle of about 85°. It is constructed of a triple back lens, a double middle, and a single front. Messrs. Ross's system is believed to differ from this, inasmuch as their objectives, from the $\frac{1}{2}$-inch upwards, are supposed to be constructed on Mr. Wenham's new formula, in which the flint concave of a triple middle is made to correct the aberrations of the anterior and posterior crown lenses. An enlarged section of this formula is shown in Fig. 31.

FIG. 30.

Quarter-inch objectives may be obtained of as high an air angle as 140°, very good for the experienced microscopist but of limited use to the student, as such glasses focus very close to the object.

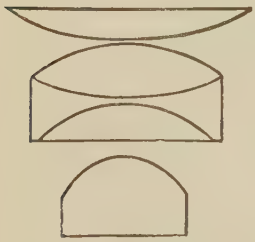


FIG. 31.

All objectives possessing a numerical aperture of more than 0·42 (50°) should be furnished with a screw collar to adjust the lenses for varying thicknesses of covering glass, and this is specially needed on Mr. Wenham's new formula. It is often a very tedious operation to find the exact adjustment requisite; but it may be approximately performed in the following manner:—

Set the collar at zero, and focus the glass upon the object; next turn the collar until the dust upon the cover-glass is in focus, when an approximate correction will have been applied; if the observer again focusses by means of the fine adjustment, the object will be found more sharply defined than before. This may only be considered a rough method. The best way is to work at some well-known

object, under varying positions of the screw collar, such as *Gyrosigma angulatum* or the Podura scale, until the best definition has been arrived at.

Under the denomination of high powers may be classed all those yielding greater amplification than the $\frac{1}{4}$-inch. They have become very common during the last few years, nearly every maker of objectives issuing an "eighth," and an immersion $\frac{1}{15}$ or $\frac{1}{16}$.

It appears probable that most of the high powers now made are constructed upon a formula similar to that last described by Mr. Wenham, and illustrated in Fig. 31 (though this is denied by some makers), as he states most distinctly that the formula is successful with all powers requiring adjustment for cover-glasses, "from the $\frac{1}{2}$-inch upwards."

Eighths and higher powers used to be specialities of the first-class makers for which very high prices were charged; but now, for a student's general work, very good eighths may be purchased for 50*s.*, of an aperture of 0·82 (110°) and good working distance.

As to magnifying power, the annexed list has been extracted from the catalogue of Messrs. Powell and Lealand, and will serve to show what degree of amplification is yielded by the various objectives, in combination with different oculars, at the recognised distance of 10 inches.

Objectives.	A.	B.	C.	D.	E.
inch.					
4	12	18	25	50	75
2	25	37	50	100	150
1	50	74	100	200	300
$\frac{1}{2}$	100	148	200	400	600
$\frac{1}{4}$	200	296	400	800	1,200
$\frac{1}{8}$	400	592	800	1,600	2,400
$\frac{1}{16}$	800	1,184	1,600	3,200	4,800
$\frac{1}{25}$	1,250	1,850	2,500	5,000	7,500
$\frac{1}{50}$	2,500	3,700	5,000	10,000	15,000

It may be useful also to add a list of the magnifying power of the oculars and objectives of Hartnack (now Prazmowski) and of Zeiss. They are used in this country to some extent, and the users always have the foible of omitting the number of diameters to which the object has been magnified, adding, however, the numbers of both ocular and objective, to which the student generally has no reference. It should not be forgotten that the amplifying powers of the foreign objectives are calculated for a tube of 153 millimetres (6 inches) in length, and most of these objectives lose in their performance if employed in the ordinary English length of tube.

Prazmowski (Hartnack) gives the following values for his oculars and objectives :—

Objectives.	Oculars.						Equivalent Focus in Inches.
	1	2	3	4	5	6	
1	15	20	25	..	..	..	2
2	25	30	45	..	..	..	1
3	50	60	80	120	..	..	$\frac{3}{4}$
4	60	70	90	140	..	..	$\frac{1}{2}$
5	100	125	160	240	..	..	$\frac{1}{4}$
6	150	180	240	350	..	..	$\frac{1}{6}$
7	200	240	300	450	600	700	$\frac{1}{8}$
8	250	300	400	600	800	1000	$\frac{1}{10}$
9	350	400	550	860	1100	1400	$\frac{1}{11}$

Immersion Objectives with Correction.

9	410	480	630	950	1300	1500	$\frac{1}{12}$
10	520	600	750	1100	1500	1800	$\frac{1}{16}$
13	820	950	1170	1700	2370	3100	$\frac{1}{25}$
15	1040	1200	1500	2200	3000	3600	$\frac{1}{33}$
18	1560	1800	2250	3300	4500	5400	$\frac{1}{50}$

Zeiss, whose objectives have been much in request during the past year or more, is noted for great working distance; his angles are low and he designates his objectives by

letters, but gives also the equivalent focal lengths in inches and millimetres; they are as follow:—

	Mark.	Equivalent Focal Length.		Angular Aperture.	Oculars.				
					1	2	3	4	5
		inch.	mm.	°					
Dry Objectives.	aa	1	25	24	20	27	36	52	70
	A	3/5	15	24	40	55	75	105	140
	AA	3/5	15	36					
	B	2/5	10	40	70	100	135	180	240
	BB	2/5	10	60					
	C	1/4	6·5	50	110	145	195	260	370
	CC	1/4	6·5	90					
	D	1/6	4·2	75	175	235	320	440	600
	DD	1/6	4·2	105					
	E	1/9	2·8	110	260	350	480	660	900
	F	1/14	1·8	110	410	550	750	1020	1390
Immersion Objectives.	G	1/8	3·0	108° water-angle	250	340	450	620	840
	H	1/11	2·3		320	440	590	800	1100
	J	1/15	1·7		440	590	800	1090	1500
	K	1/20	1·3		590	790	1060	1450	1980
	L	1/25	1·0		760	1030	1380	1890	2580
	M	1/33	0·75		1010	1360	1840	2520	3450
	Equivalent focus of oculars, mm.				48	42	30	24	18
	Do. do. in inches ..				1·88	1·64	1·18	·94	·70

In the foregoing remarks it will be observed that moderate angles have been advocated *for students' use;* but it must by no means be inferred that moderate angles are the best. The higher the numerical aperture of an objective, the nearer does the front lens approach the object when in focus, and this has been urged as an objection against the use of large apertures. There is some force in this objection—in dissecting, for instance, it is absolutely necessary to have room underneath the objective for the use of the various knives, needles, scissors, and other instruments, which one cannot get with large apertures; but when the object has been prepared there is no valid reason why the best lenses should not be used for its examination.

Much has been said relative to the aperture of objectives; therefore it may be of interest to the student to know how the measurement of this angle may be set about. Colonel Woodward and Mr. Wenham both prefer modifications of Professor Robinson's method, as being the most accurate.

Colonel Woodward screws the objective to be tested into a tube, which pierces the shutter of his dark room, the back of the objective being towards the light. A parallel pencil of sunlight is then thrown through it by means of a solar mirror, and the cone of light is measured off directly upon a cardboard protractor, arranged with its centre in the focal point of the lens.

Another method is to fix the microscope body so that it may rotate upon a pin placed with the focal point of the objective as a centre. This is then placed on a board graduated into degrees, and the light of a candle, placed a few feet off, viewed through the eye-piece. The body of the microscope is turned until only half the field remains illuminated, and the angle read off upon the graduated circle below. Mr. Wenham declares this method faulty, and also passes some strictures upon Colonel Woodward's plan, stating that a border of diffused light exists which renders the measurements inaccurate, the extreme angles having often to be guessed at.

Dr. G. E. Blackham, in his paper already quoted, gives a method suitable for most objectives, a plan which can easily be carried out with one of the stands fitted with a swinging substage. A plano-convex lens is employed, of such thickness that when the plane side is connected with the under surface of the object-slide with softened balsam, the thickness of slide, balsam, and lens shall equal the radius of curvature. This lens is connected by balsam to the under side of the slide containing a balsam-mounted object.

Dr. Blackham's lens is of crown glass, with a thickness of 0·33 inch, and a radius of curvature of 0·45 inch.

The swinging substage bar is made to carry a small candlestick, in which is placed a candle, so that when the body of the microscope is horizontal, the light can be swung round the object as a centre until either the image of the object is imperfect or the centre of the field darkened.

In practice, the object (with the plano-convex lens attached to the under surface of the slide) is laid upon the stage and exactly focussed, the best definition being obtained by use of the collar adjustment. If now the angle that the most oblique ray makes with the optic axis be measured, the angle of deviation in glass is obtained, from which the *numerical aperture* can be obtained by simple inspection from the table given in Chapter V., and the air or water angle quite as readily estimated.

Professor Abbe's apertometer will be described in Chapter IX.

CHAPTER IV.

ACCESSORIES.

HAVING made the reader familiar with the compound microscope, its eye-pieces and objectives, we must now consider those appliances which conduce to excellence in microscopical examinations, or which render the work more easy and more quickly performed. It must not be imagined, however, that the possession of all the following accessories is imperative for microscopical studies; far from it, as precisely the same effect as that produced by the most costly accessory can often be obtained (with more trouble) by very simple and inexpensive means.

FIG. 32.

In the first place, we may consider the draw-tube, which is shown in Fig. 32. It is supplied of considerable length to most monocular microscopes, being cut down in many binoculars.

Into this draw-tube the eye-pieces should be fitted, and the former then becomes a means of increasing the amplifying power of the combination. By simply drawing out the tube the object is thrown out of focus, and the objective has to be carried nearer the object. This wonderfully alters the relative distances of eye-piece and objective from

the object (conjugate foci), and the result is a considerable increase in amplification. It must not be forgotten, however, that a limit will soon be found to this method of increasing the magnifying power. The longer the tube, the more of the periphery of the field will be cut off; and further, the aberrations, which have been fairly corrected for a moderate or short tube, may no longer obtain correction when the body of the microscope is unduly lengthened.

Some of these tubes are plain, while others are divided into inches and parts, so that results once obtained can be recorded, if necessary.

There are many uses for the draw-tube, which the student will be sure to find out for himself, and he will find that if it is graduated, as shown in the figure, his work will often be rendered much more easy. Some of its uses will be found described under the subjects of Dissecting and Micrometry.

FIG. 33.

Into the lower end of the draw-tube the erector (Fig. 33) is made to screw. It consists of a tube about 3 inches long, having a meniscus (concavo-convex) at one end and a plano-convex at the other, a diaphragm being placed about midway between them. The convex side of each lens is turned towards the eye-piece, this combination producing a second inversion of the image, so that it is seen in its natural position. This is of great use to the tyro dissector, as he has then but little difficulty in the use of his dissecting instruments; but if any one who has learned to dissect without it should attempt to use the erector, he will find it is quite as hard to unlearn as to learn.

The erector, when screwed into the lower end of the draw-tube, enables the observer to employ a greater range

of magnifying power without changing the objective, and this is especially useful in either dissecting or photography by means of the microscope.

The field of view is also much increased, so that a very large object can be included when using the half-inch objective, and by pulling out the draw-tube (containing the erector, of course) a very considerable increase in amplification is obtained; thus, in the author's dissecting microscope, when the erector is placed as near to the objective as possible, the magnification obtained scarcely exceeds 8 diameters, while when the draw-tube is pulled fully out the amplification is increased to 130 diameters. It should be understood that the defining power of a microscope is not *increased* by the addition of an erector.

The double nose-piece next claims our attention, and was devised by Mr. Brooke for the purpose of quickly changing

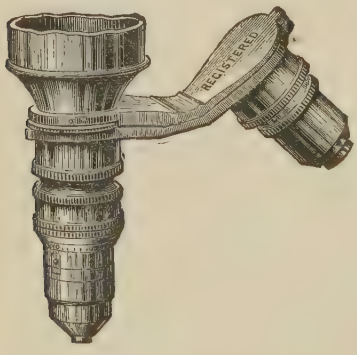


FIG. 34.

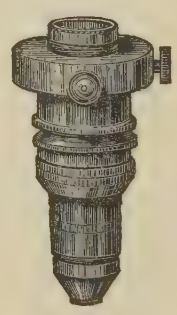


FIG. 35.

the objectives without the trouble of unscrewing and screwing up again each time. This accessory is made in several forms, but that shown in Fig. 34 is specially recommended, the straight pattern being sometimes inconvenient, owing to the one objective touching the stage before the other required for use is in focus. It is a very useful addition to the

microscope, and should be purchased by all who can afford it, when much work has to be done; but on no account should the beginner purchase one of those made to hold three or four objectives—they are too heavy, and often nullify completely the action of the fine adjustment.

Some years ago Mr. Swift introduced a centering nose-piece, which is shown in Fig. 35. In this accessory the objective can be accurately centred after it has been screwed into the nose-piece, which latter screws into the lower end of the body of the microscope. The centering screws are similar to and act in the same manner as those connected with the achromatic condenser. Centering nose-pieces are not very common; the author has seen but one in use, and that with high powers only.

Fig. 36.

The student will find the bull's-eye condenser (Fig. 36), or stand condenser, as it is sometimes called, a necessary adjunct, an instrument which may be used in a variety of ways; but its proper use, and the methods of illumination generally, can only be completely understood by consulting a few diagrams of mirrors and lenses which

show their action upon various rays of light. An illustration of the bull's-eye condenser is given at Fig. 36, from which it may be understood that it is really a plano-convex lens of crown glass, but mounted in various ways, and the student should remember, on purchasing one, to select such a form as may be turned and twisted in every desirable direction.

There are many uses for the bull's-eye condenser. It is an indispensable instrument for the illumination of opaque objects; with it the microscopist is enabled to throw parallel rays on to his mirror from the lamp; he may use it as a spot lens by fixing a small disc of dead-black paper on the flat surface of the lens, and as an Amici prism for the resolution of diatoms, Nobert's test lines, &c.

A smaller condenser, generally called a "condensing lens" or "stage condenser" is supplied with some microscopes, but its use is very limited, and the student is advised to purchase only the independent stand condenser. With the higher powers a more intense illumination can be obtained by a combination of two condensers, the correct method of using which may be made out by inspection of the diagrams in the next chapter.

As to quantity of light, we have always found that students have too much of it, and therefore it may be as well to consider the diaphragm next. The diaphragm is a thin plate of metal perforated with holes of various shapes and sizes, and is used in order to cut off the superfluous rays from an object, or leading to an object, under examination. At one time they used to be placed at considerable distances beneath the object, but the practice nowadays is to put them immediately beneath the slide, which doubtless is their proper place.

The ordinary form is shown at Fig. 37, and is called a wheel of diaphragms; it is usually supplied to all student's and third-class instruments.

It is made of many degrees of excellence, and therefore every cheap instrument purchased should be specially examined with a low power (3 or 4-inch objective), to see

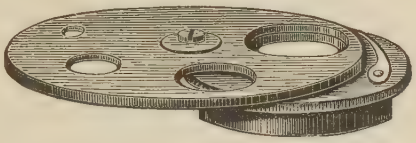


FIG. 37.

whether the apertures are concentric with the axis of the instrument. Every part of the diaphragm should be well blackened, in order to prevent the presence of reflected light, which would interfere with the illumination.

Collin's graduating diaphragm, illustrated by Fig. 38, was the first of its kind. It consists of four movable shutters, acted upon by a lever, so that the whole of them

FIG. 38.

FIG. 39.

may be moved inwards or outwards simultaneously. The opening thus produced is not circular, but this is of little importance in actual working.

The graduating or "iris" diaphragm of Messrs. Beck is illustrated in Fig. 39. It differs from that of Mr. Collins only in the number of the shutters, which thereby produce a nearly circular aperture. Mr. George Wale, of New

York, supplies a new form of iris diaphragm to all his "working" microscopes.

It is now universally appreciated that the diaphragm must be placed immediately beneath the object, in order to get the best results. This has been attained by the "calotte" diaphragms now made by Messrs. Zeiss, Swift, and others. This form is placed *over* the achromatic condenser, on a level with the stage, as shown in Fig. 40; but the simplest way to effect this is by using a thin plate of aluminium 3-inch by 1-inch, with the desired aperture pierced in its centre, and furnished with two spring clips, so that the slide may be placed upon it in any required position, such as has been used by the author for the past seven years.

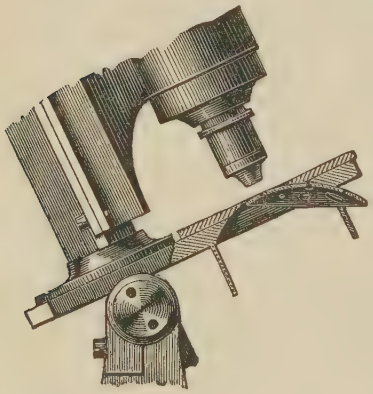


FIG. 40.

There is another method of excluding the excess of extraneous light, used by the author since 1873 with very good results—viz. that of slipping over the objective a small perforated cardboard and aluminium nozzle, blackened inside, the central hole in the front being just a trifle less than suffices to admit the rays passing from the field of vision.

For many purposes it is necessary to concentrate the light to a greater extent than can be done with a mirror alone, and in such instances an achromatic condenser is usually employed. This may be improvised from an ordinary objective made to screw into an adapter below

the stage, a $\frac{1}{2}$-inch or $\frac{4}{10}$-inch objective of wide angle forming a very useful condenser.

A substage condenser consisting of a plano-convex lens of $\frac{3}{4}$-inch focal length was used by Wollaston for illuminating the object upon the stage; this was improved by Goring, but still did not satisfy the requirements of microscopists. In 1840 the achromatic condenser was introduced, and for many years afterwards continued to be made as a low-angle objective, adapted in various ways to the stage or substage. Whether achromatic light for illuminating purposes is absolutely necessary is a question we cannot discuss here.

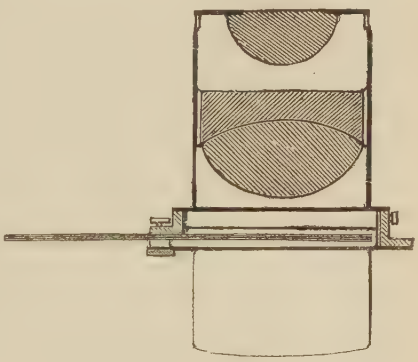


FIG. 41.

Messrs. Beck, Ross, and Powell and Lealand, together with other makers, have produced at various intervals modifications of the achromatic condenser; but a departure from the usual custom was made in 1865 by Mr. Webster, who communicated a drawing of his instrument to the pages of 'Science-Gossip,' and which may be seen in Fig. 41. The diaphragm with which it was furnished may be seen in Fig. 42.

Mr. Collins's "Webster" condenser is shown in Fig. 43, and is a form extremely well-liked by all practical microscopists; it is made so that the diaphragm may be carried quite close to the lens, and this can be further improved, when dark-ground illumination is required, by placing a circular piece of black paper on the lens itself. It can be used with ease in combination with the polariscope.

The use of the condenser may be demonstrated in various ways, but when employed in connection with the diaphragm, one of its uses may be shown by taking a slide

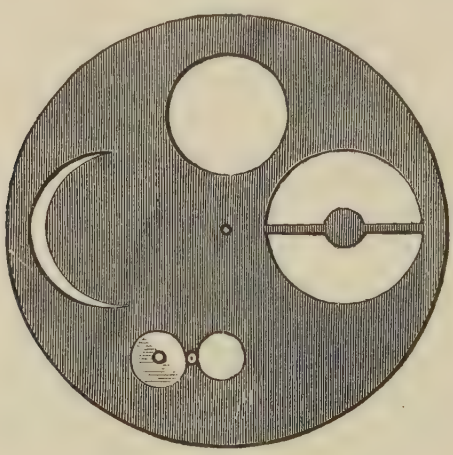


FIG. 42.

of *G. angulatum*, and using the condenser with a small central aperture. In this way it will be found very difficult indeed to resolve the markings properly, but if the central light be stopped out, so as to utilise the oblique rays only, the same effect will be produced as by swinging the mirror round to the side and getting obliquity in that way.

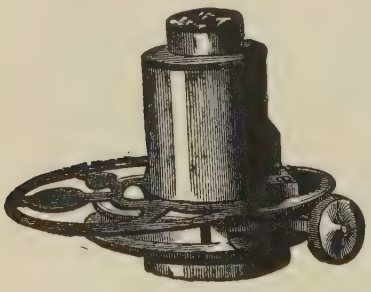


FIG. 43.

In using the achromatic condenser, great care must be taken to insure the coincidence of the optic axis with that of the microscope, by manipulation of the two adjusting screws. This is most easily accomplished by the use of the centering

glass, made and sold by most opticians. The complete condenser of Messrs. Swift and Son is a very ingenious piece of apparatus; it is made to supply the place of a substage with all the ordinary compound substage apparatus. It comprises an achromatic condenser with an aperture of 140°, a spot lens, a contracting diaphragm, a revolving diaphragm and set of stops, polarising prism, together with revolving mica and selenite films. The polariscope is mounted on an eccentric arm, so that it can be thrown in and out of use instantly, which is an important feature, for it may be presumed that the polariscope would be much oftener employed if it could be put in and out of action as easily as shown in Fig. 44.

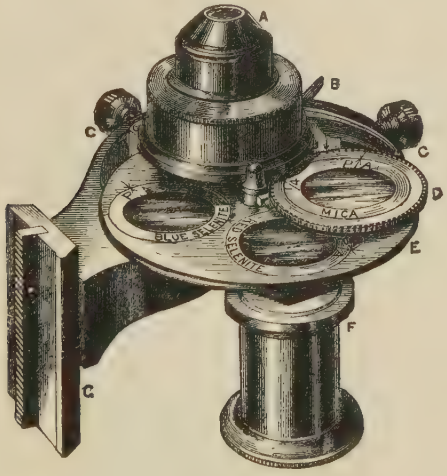


FIG. 44.

In the microscope itself Mr. Swift now inserts the analysing prism of the polariscope in a brass box over the Wenham prism, thus facilitating its use, as there is no unscrewing of parts to fix the analyser in position.

Reade's double hemispherical condenser consists of a

plano-convex lens about 1½ inches in diameter, its flat side being placed next to the object, and surmounted by a smaller lens of the same form (hemispherical). A very oblique illumination is possible with this condenser, and as made by Messrs. Ross and Co. it is shown in Fig. 45.

Several makers are now producing oil-immersion condensers, in which the under side of the slide is connected with the condensing lens by means of a film of castor oil, oil of cedar wood, or glycerine. That made by Messrs.

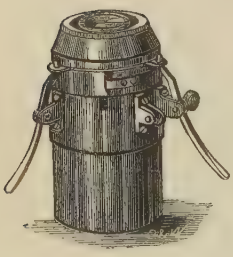


FIG. 45.

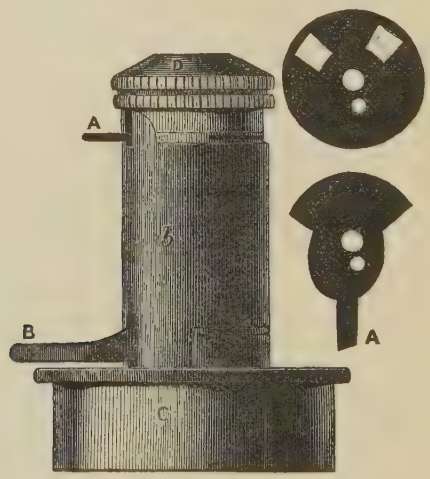


FIG. 46.

Powell and Lealand is shown in Fig. 46. It is not achromatic, but is able to utilise rays of 130° balsam angle. Beneath the lens are fitted diaphragms of peculiar pattern, and by its use the most difficult test objects may be resolved with the mirror in the optic axis of the microscope.

Messrs. Watson and Sons of Holborn produce an oil-immersion condenser, in which the lens is mounted on a plate of ebonite; the object slide being placed above it, the latter is held in position by a pair of clips, and is then ready for examination.

The swinging substages of Messrs. Ross and Beck are

useful for obtaining very oblique rays of light with the help of the condenser. This has been accomplished in another way by Messrs. Swift and Son, in their radial traversing substage illuminator. While regretting that this firm could not have found a shorter title for this accessory, we must say it seems more generally applicable than most other modes of producing obliquity of light. It is shown in Fig. 47.

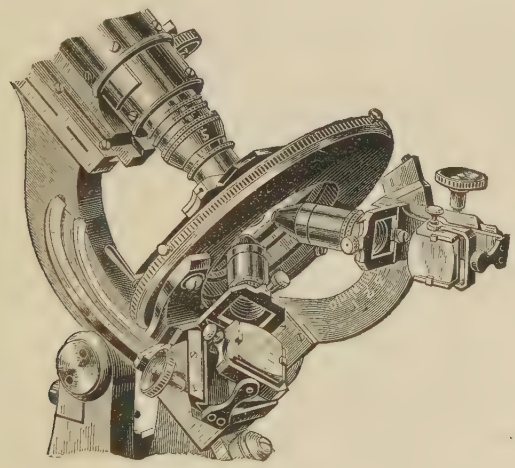


FIG. 47.

This apparatus has been constructed for the purpose of increasing the resolving property of high-power objectives by causing still more oblique pencils to impinge upon the object than can be obtained by most methods. The arrangement consists first of an arc-piece, fixed below the stage, radial to an imaginary line drawn through the axis of the microscope objective, in the same plane with the object. On this an achromatic condenser of special construction is made to travel, thus keeping the rays of light on the object during its entire traversing. These rays converge and terminate in a focus through the front lens, in a highly concentrated form. The condenser is illuminated

by a rectangular prism, for condensing light into the achromatic combination. The next part consists of a second arc-piece placed at right angles to the former one; this also carries a similar achromatic condenser and illuminating prism, which move radial to the centre. Both these arc-pieces are so divided that each pencil of light can be projected at a similar angle, and previous results can always be recorded in the same way. Difficult test objects are readily resolved, especially such diatoms as have rectangular striæ or markings. With a $\frac{1}{4}$-inch objective, the diatom *Navicula rhomboides* is easily resolved into squares. The markings on *G. angulatum* by the same means are made to stand out in bold relief like half spheres. Those usually considered easily resolvable only require one pencil of light to show the markings. When this is the case, the

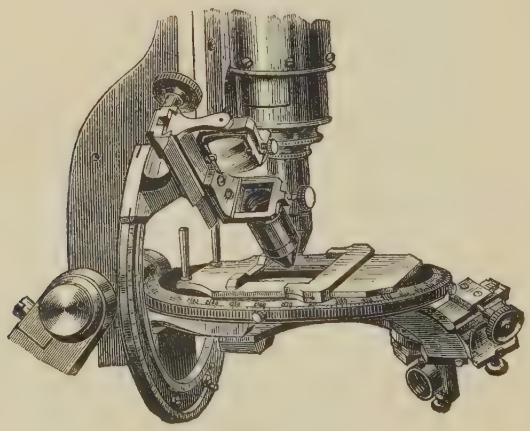


FIG. 48.

rectangular arc-piece with all its illuminating apparatus can be turned away from the microscope stage as shown in Fig. 48. The same illustration shows how opaque objects may be illuminated, viz. by moving the condenser of the first arc-piece above the stage of the microscope, when a

pencil of light can be projected on to the object more perpendicularly than with the bull's-eye condenser, thus preventing shadows in coarse or deep objects which often produce distortion and false appearances. When the apparatus is used for opaque objects with a lower power than the 1-inch objective, the achromatic combination can be removed and the light directed from the prism, which can be made to give convergent rays sufficient for use with a 4-inch objective.

Wenham's reflex illuminator is valuable for use with high powers; but it possesses the disadvantage that all objects for use with it must either be selected or specially mounted. It has been described by the designer at some length in the seventh volume of the 'Monthly Microscopical Journal,' p. 237, in which he states that by its use the markings on *A. pellucida* were brought out by an eighth objective, which had never shown them before. Fig. 49 gives a sectional diagram of the illuminator, which has been manipulated from Mr. Wenham's sketch. In using this accessory, the glass cylinder is brought up level with the stage, the centre of rotation is set truly by a dot on the fitting, as seen with a low power. A drop of water is now placed on the top of the cylinder, upon which the slide is laid. The effect on the Podura scale is superb, the whole surface appearing dotted with bright blue spots in a zigzag direction. The plane mirror is the best for throwing up the light, and when the finest effect has been obtained

FIG. 49.

upon any object, by simple rotation of the illuminator, an exquisite unfolding of structure takes place. Objects *on the slide* are brilliantly illuminated, while those on the cover are nearly invisible.

Another illuminator has been devised by Mr. Wenham, and is shown in Fig. 50. It looks very much like the half of a broken button, which, nevertheless, collects and concentrates a surprising amount of light. It consists of a semicircular disc of glass $\frac{1}{4}$ inch in radius, the edge being well rounded and polished to a transverse radius of $\frac{1}{10}$ inch. This may be obtained from Messrs. Ross and Co. and Messrs. Baker.

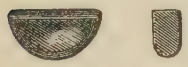


Fig. 50.

We have heard from several microscopists that they have been unable to use this illuminator successfully, and it has generally been the case where it has been mounted for use in the substage. The best plan is to purchase the article *unmounted*, and to make it adhere, for use, to the underside of the slide, with a little glycerine and gum. This illuminator does not require the addition of any condensing lens, the necessary obliquity of light being readily obtained by swinging the mirror to the side. Mr. Wenham, in writing to the 'English Mechanic,' says that while he was never successful in the patient manipulation required to bring out the striæ of *Amphipleura pellucida*, yet with the new illuminator he succeeded at once, and on every subsequent occasion.

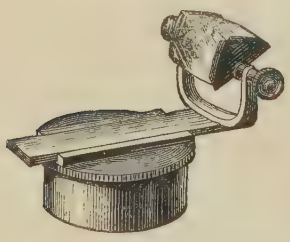


Fig. 51.

The Amici prism ranks next. It is made to exhibit one plane and two lenticular surfaces, thus concentrating and reflecting the rays at the same time. It is mounted so that it may be

used in the substage, as shown in Fig. 51, or better perhaps upon a separate stand, so that it is entirely independent of the microscope or stage, Fig. 52.

In using this prism it is set beneath the stage at such a distance from the axis of the microscope as to furnish rays of sufficient obliquity, and in order to produce the best effect the stage should be capable of rotating round the fixed luminous rays, as with line objects, such as the silicious valves of diatoms, the *Navicula rhomboides*, for instance, there is nearly always one position in which the markings are shown to the best advantage.

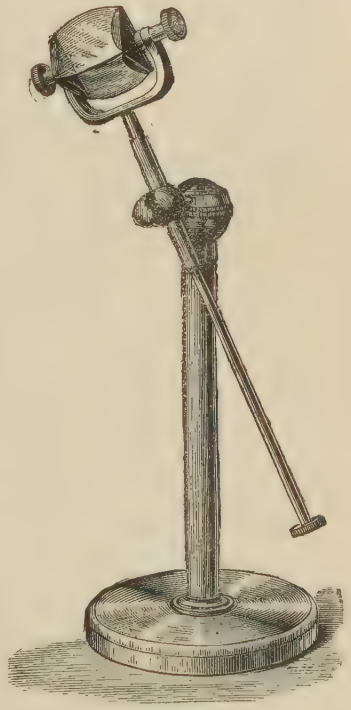


FIG. 52.

Colonel Woodward's prism has been described upon p. 246, vol. i. of the 'Journal of the Royal Microscopical Society.' It consists of a right-angle prism of crown glass, the long side of which measures $\frac{3}{4}$ inch by $\frac{1}{2}$ inch wide; it is cemented into a base-piece of brass, and supported on a stout steel rod three or four inches long. Colonel Woodward tells us that the whole apparatus ought not to cost more than three or four shillings, but we are afraid he has not consulted the optician in this matter. To use it, the steel rod is slipped into a dark-well holder, and putting a drop of oil on the upper face of the prism, it is placed in contact with the under surface of the slide.

The light of a paraffin lamp is then to be condensed upon the object through one of the faces of the prism, until it is seen by inspection through the other face that the objective is well illuminated.

Nachet's prism is shown in Fig. 53, the upper and lower surfaces of which are convex, by reason of which the rays of light proceeding from the mirror are conveyed by the lower surface, and after undergoing two reflections are finally brought to a focus upon the object. The prism should be so mounted that it may be revolved for the same reasons as have been already given when treating of the Amici prism. The Nachet prism is not much used in this country, and in its present form it is constructed with the lower surface plane and not convex as in the original. Then, again, there is the right-angle prism which finds occasional use. The proportion of light reflected by a prism is greater than can be obtained from the ordinary plane silvered mirror, which always yields secondary reflections from the surfaces of the glass.

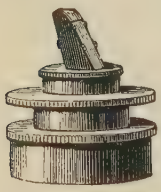


FIG. 53.

Lord Osborne's illuminator or diatom exhibitor is made by Messrs. Baker, of Holborn, and is much praised by many microscopists as an aid in resolving markings on plane surfaces; it is somewhat similar to Reade's condenser in the form of the lenses, but they are mounted differently.

The subject of condensers and prisms is one which should be well studied by the beginner before he decides to purchase; always talk the matter over well with experts before committing yourself to any particular form.

The illumination of opaque objects under high powers has been attempted by many, but the most successful appliances for this purpose are easily enumerated—Powell and Lealand's, and Beck's vertical illuminators, and Tolles'

interior illuminator. The best objects to show the method of using these accessories the student will find to be the Podura scale and the diatom *Navicula rhomboides*, or the *Amphipleura pellucida*, which will be illustrated in the next chapter.

Messrs. Powell and Lealand's vertical illuminator consists of a highly polished plate of glass, set at an angle of 45° in an adapter which is screwed on over the objective. There is an aperture in the side furnished with a revolving diaphragm, through which the illuminating rays are made to pass. These rays are reflected downwards, through the objective, upon the object, as may be more easily understood on reference to Fig. 54 *a* and *b*.

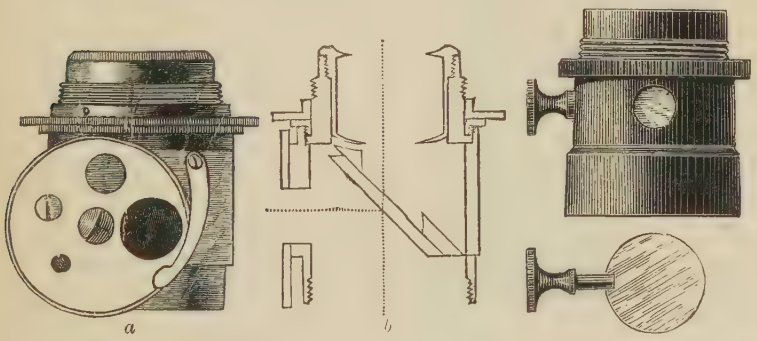


FIG. 54. FIG. 55.

Beck's vertical illuminator is very similar to the above, but the plate of glass is replaced by a thin glass disc or thin cover attached to a milled head, as shown in Fig. 55. This seems to be a more handy form than the first, and for ordinary instruments is perhaps the best. Still the author must admit that with Messrs. Powell and Lealand's workmanship their form of vertical illuminator is all that can be desired. In December 1880, Mr. Powell exhibited at a "Scientific evening" of the Royal Microscopical Society *Amphipleura pellucida* lighted up with this illuminator,

and under their new twelfth objective the result was superb.

This method of vertical illumination does not answer so well with covered objects mounted dry; there is a mistiness produced by the reflection from the covering glass, while with objects mounted in balsam the illumination is perfectly satisfactory.

The illuminator made by Tolles, and called the "interior illuminator," may be seen in enlarged section at Fig. 56. A prism is inserted between the combination of the objective, which collects and throws down the light through the objective on to the object below. The curves of the lenses figured form that celebrated $\frac{1}{8}$ upon which there was so much controversy as to the real aperture some few years since.

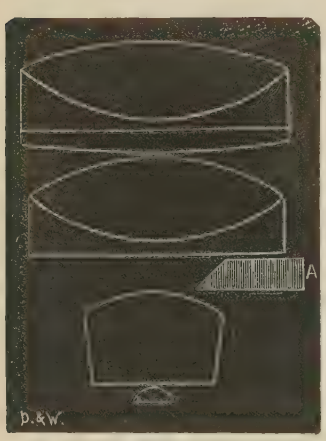


FIG. 56.

Another simple method for illumination under high powers was described by Mr. James Smith at the March meeting of the Royal Microscopical Society, 1880, extremely well suitable to such objects as Podura scales, diatoms, &c., under powers as high as the $\frac{1}{16}$.

Mr. Smith uses the bull's-eye condenser only, and places it with the plane side uppermost, just above the stage, the lamp being set in front at a distance of two or three inches. The light enters the condenser, and is reflected very obliquely upon the slide from the plane surface of the bull's eye.

An appliance often used to effect dark-ground illumina-

tion is the spot lens: it is of very simple construction, being a plano-convex lens, upon the upper flat surface of which is drawn a circular spot of black varnish, so as to exclude all central rays and prevent them passing into the objective. It is shown in Fig. 57.

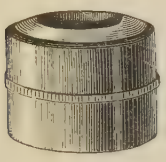


FIG. 57.

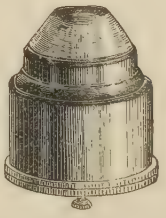


FIG. 58.

In the best instruments the paraboloid is the dark-ground illuminator, as rays of greater obliquity can be obtained by the use of this accessory than by the spot lens. It is shown in Fig. 58.

In using the paraboloid the plane or flat mirror must be used to throw parallel rays upwards, and these are reflected from the internal surfaces of the glass at such an angle that with objectives of moderate aperture the field appears quite dark, the rays simply illuminating the object but not entering the object glass. The track of these rays may be seen on reference to Fig. 59.

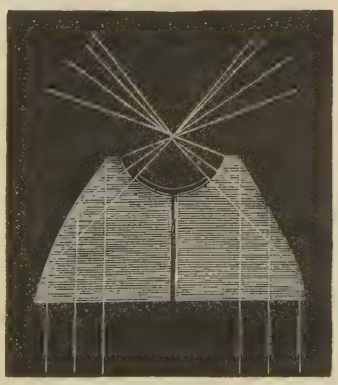


FIG. 59.

A central stop or dark well is placed in the hollow of the paraboloid, and is capable of motion either upwards or downwards by means of a pin which runs through a hole drilled in the glass;

when this is pushed upwards it cuts off the less divergent rays which would otherwise proceed from the apparatus. When lamplight is used, the bull's-eye condenser should be interposed between the lamp and the plane mirror, in order to parallelise the rays which fall upon the latter. A large mirror is requisite for the proper manipulation of the paraboloid, and this is not usually obtained in third-class instruments.

When objectives of wide angle are used, a somewhat different arrangement is necessary to secure dark-ground illumination. In the 'Monthly Microscopical Journal' for July 1869, Mr. Wenham reproduces the illuminator which he first described in 1856 as "A method of illuminating opaque objects under the highest powers of the Microscope." Fig. 60 will illustrate the subject of these remarks. Cemented to a glass slide with Canada-balsam is a nearly hemispherical lens "with a segment removed so as to leave the thickness equal to about one-third the diameter of the sphere. The circle formed by the removal of the segment is blackened in order to exclude all rays below the incident angle of total reflection. This lens is intended to be used in conjunction with the paraboloid as shown in Fig. 60. The rays pass through the lens in a radial direction without refraction, and proceed till they reach the upper surface of the thin glass cover, where they are totally reflected upon the object."

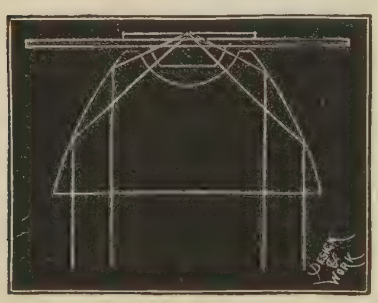


FIG. 60.

An immersion paraboloid has since been devised by Dr. Edmunds, and made by Messrs. Powell and Lealand; it is somewhat similar to the above in principle.

For the illumination of opaque objects several kinds of apparatus are used, and of these we may consider first, the parabolic reflector as made by several opticians. This is most useful for low powers, from the 2-inch to the $\frac{2}{3}$, and is made to fasten on to the objectives by means of a spring clip, so that it may slide up or down or be turned round to secure the best illumination.

When this reflector is used with a lamp, the bull's-eye condenser must be placed between them with the flat surface towards the lamp, and its focal point so adjusted that the rays emitted from the bull's-eye shall be parallel. The chief use of this reflector is to reflect side light from all points of its surface to a point situated on the plane of the base in the focus of the objective. The form as made by Messrs. Beck is illustrated in Fig. 61.

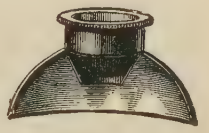


FIG. 61.

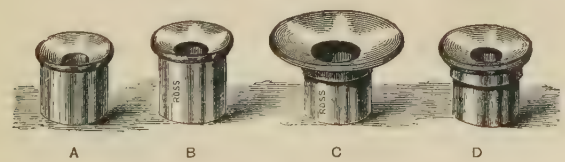


FIG. 62.

At A, B, C, and D, Fig. 62, are shown the accessories called Lieberkühns, or Lieberkuhn's reflectors, which are used occasionally for the vertical illumination of opaque objects. They are small silvered concave mirrors mounted on short tubes, to admit of adjustment on the objective, and thereby yielding the maximum illumination. A separate Lieberkuhn is required for each object-glass, as the focus of the concave mirror has to be adjusted to that of the objective.

In using this appliance, parallel rays of light are thrown

through the stage by means of the plane mirror, which on meeting the Lieberkuhn are deflected upon the object, as shown in Fig. 63. Lieberkuhns are not much used at the present time, and can only be applied to objects which are so mounted as to allow of the passage of marginal rays through the slide.

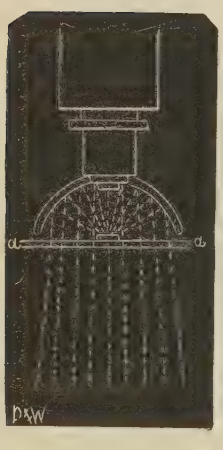


FIG. 63.

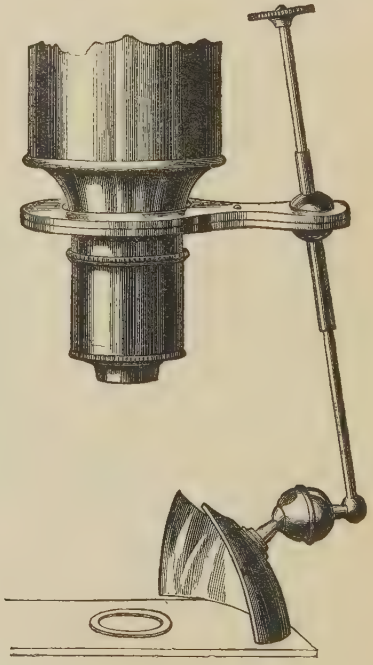


FIG. 64.

Side reflectors, which are much used for the illumination of opaque objects, are either mounted in the same manner as stage-forceps, or as shown in Fig. 64, in a collar which spans above the objective, as made by Mr. Swift.

The side reflector may be used to illuminate opaque objects on the stage very successfully, and with such obliquity that many delicate markings which are unobserved

with the Lieberkuhn are brought out without difficulty. In use, the bull's-eye should be interposed between the reflector and lamp, to render the light parallel, exactly the same as with the parabolic reflector.

When the object is mounted on an ordinary slide, as a transparent object, a dark ground may be produced by using one of Lister's dark wells, which are merely blackened stops, their deep cup-shape insuring perfect blackness, their diameter intercepting the rays which would otherwise pass into the objective. The engraving (Fig. 65) is an illustration of a dark well in its sub-stage mounting, as made by Messrs. Beck.

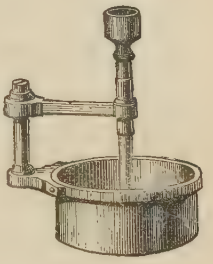


FIG. 65.

For these dark wells the author often uses small circles of black paper mounted on the ordinary 3-inch by 1-inch slips, and for most purposes they answer admirably.

We now come to several very simple but important additions to every microscope, the first of which is generally supplied with all instruments. Fig. 66 delineates the stage forceps, shown holding a fly for rough examination under

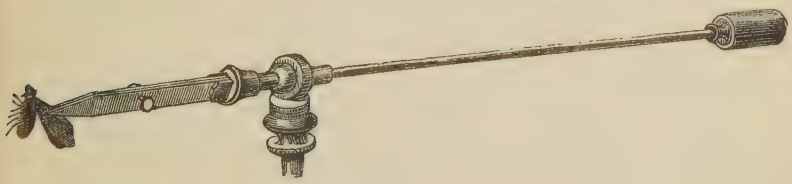


FIG. 66.

low powers. They are useful chiefly to beginners, with such objectives as the 3-inch or 2-inch, but are altogether unsuitable for high powers.

In the investigation of minerals it is often necessary to examine small angular pieces which require to be viewed on

all sides. In order that this may be done easily, Messrs. Beck make what is called a stage mineral-holder (Fig. 67), one of the jaws being movable in a right line, so that it may clamp any sized specimen, and by turning the milled

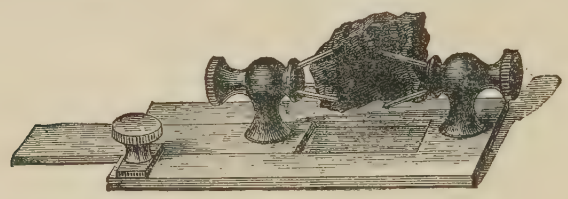


Fig. 67.

head of the jaw the mineral is made to revolve. Fig. 67 will perhaps show more clearly the action of this holder than any description can do.

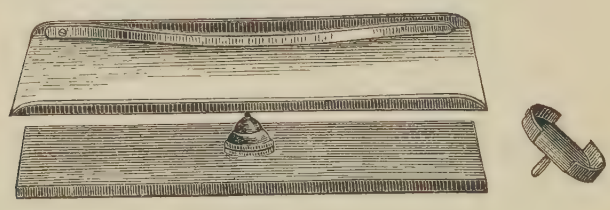


Fig. 68.

Morris's rotating stage (Fig. 68) often serves the purpose of stage forceps. In its improved form it can be used for both opaque and transparent objects. Small flies, larvæ, beetles, &c., can be affixed to the cork by means of a small pin, or with gum, and as the stage moves upon a secondary plate by means of a ball-and-socket joint, the object can be placed in a variety of positions hardly possible by any other means except the disc-holder of Messrs. Beck.

Beck's disc-holder, shown at Fig. 69, is for the purpose of holding for examination under the microscope the small discs upon which objects have been temporarily or per-

manently mounted. The object is attached to the disc by the aid of gum, or any other suitable adhesive material, and when placed in the holder for observation, the disc can be rotated in both vertical and horizontal directions. These discs are very useful for many objects, especially those not needing a cover, and Messrs. Beck supply boxes into which they are fitted when not in use, so that they may be kept excluded from the dust.

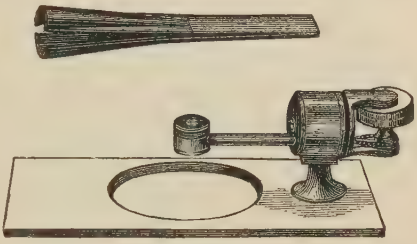


FIG. 69.

With many objects, in order to examine them minutely, all that is necessary is to place them upon a glass slip, add a drop of the right medium, and cover with a thin covering glass.

This method of working is all very well for organisms which are still, or comparatively so; but when we come to examine Rotifers, the Entomostraca, and other active forms of life, we find means are required to keep them in one position. Suppose, for instance, we wish to examine an Entomostracon, which turns out to be *Bosmina longirostris:* during its rapid motions through the water we would be apt to come to false conclusions concerning it, but if we prevent the organism from moving it can then be studied in all its details.

The simplest and least expensive way of examining the Infusoria and other moving micro-organisms is by the use of the Rotifer-trap of Mr. F. Bedwell. This consists of a few filaments of cotton wool placed upon the under glass of a "live-box." The organism contained in a drop of water is then run over it, and eventually becoming entangled

amongst the fibres, is kept comparatively still, by which its form can be clearly made out.

Two forms of cages or live-boxes are shown in Figs. 70 and 71. They are, however, not all that can be desired. Just so much pressure must be applied to the cap as is necessary to keep the animal still, and no more, or it will be crushed and distorted, so that the cap requires much dextrous manipulation. Oftentimes such objects as Entomostraca, &c., will not display themselves to advantage, so that the cap requires loosening, to be again squeezed down at an opportune moment. Moreover, it is generally found that some particular organism will get near the periphery of the cover, and in this all the interest may be concentrated, yet the objective will not reach it if it happens to be of the form of Fig. 71, and if we adopt the form of Fig. 70 we are often precluded from using the achromatic condenser to any portion but the centre of the slide. Instead of this we may use the compressorium as made by Messrs. Beck, Ross, Collins, and others, for use with high powers; but Piper's form, made by Mr. Swift, and illustrated in Fig. 72, will be found more convenient.

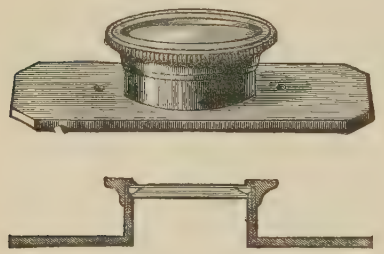


FIG. 70.

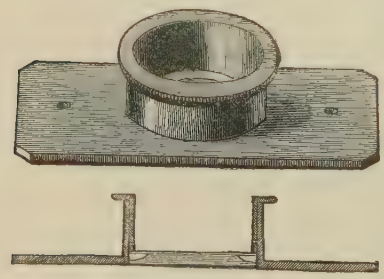


FIG. 71.

The most delicate pressure can be applied by means of

these instruments, and all such intended for real use should be reversible, so that the objects may be easily viewed from both sides, and this can be done with the form shown in Fig. 72.

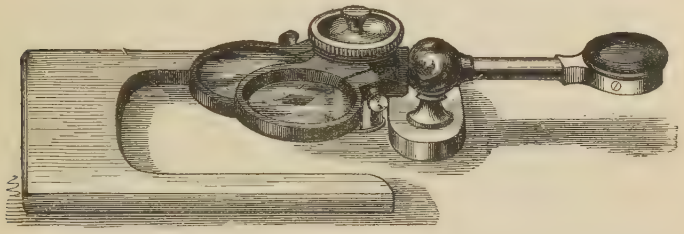


FIG. 72.

A form of compressor differing from all others has been devised by Mr. Holman, U.S.A.; the top or mica cover is fixed while the lower thicker plate of glass is raised or depressed by means of a screw-nut and spiral spring. The employment of a thin mica cover is certainly an improvement, and one which English opticians would do well to follow. In this we imagine every practical microscopist will concur, as the breakage of a glass cover in the middle of an interesting observation is, to say the least, vexing.

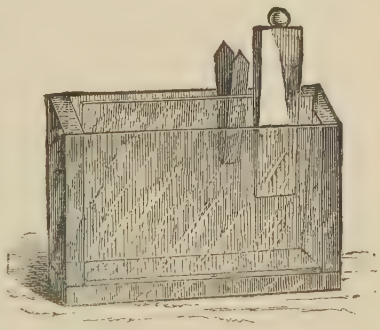


FIG. 73.

Zoophyte troughs may be easily constructed by the student; the form shown in Fig. 73 is made of a thick glass base plate and ends, while two pieces of thinner material furnish the front and back, which should be of the same thickness as the thinnest slips are cut from.

This trough should measure $\frac{6}{10}$ of an inch in width,

2 inches in length, 1½ deep at the front, and 2 inches at the back; the base plate and ends being made from glass $\frac{2}{10}$ of an inch in thickness and cemented together with marine-glue.

For higher powers, the form of trough shown in Fig. 74 is desirable. It consists of a glass slide, 3 × 1½ inches, upon which is cemented with marine glue an ebonite or glass semi-rectangular piece as shown in the figure; the half of a flat indiarubber ring will answer admirably, the front being formed of a piece of thin glass.

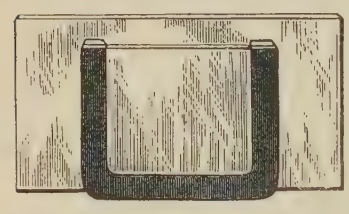


FIG. 74.

In use, nearly fill the trough with water, place it on the stage of the microscope, and incline the body of the instrument so that the observations may be made with comfort; adjust the lamp and concave mirror so that the most intense and central light is thrown through the instrument; and lastly, adjust the diaphragm until most of the marginal rays of light are cut off, there remaining only just sufficient light to work with. In all cases apply a larger aperture only when absolutely necessary.

It may be here stated that the moderate use of the microscope, either monocular or binocular, when employed in the above manner, will not injure the eyesight of a healthy person. When, however, an excessive glare of light is constantly employed, the eye becomes less sensitive to ordinary light; excess of illumination is a common fault with beginners.

The several forms of zoophyte troughs such as those shown in Figs. 73 and 74 may be obtained from any maker or dealer in microscopic apparatus.

Fig. 73 is a wide trough, but it may be narrowed by means of the wedge and spring, which drives a thin glass

partition close to the front plate; Fig. 74 is not adjustable, but is easily made by the student.

Another adjustable form of trough is Botterill's, which consists of two brass or ebonite plates bolted together, as shown in Fig. 75, the plates of glass being separated, according to the space required, by an ordinary indiarubber ring of the requisite thickness. The trough can thus be taken apart and the glasses cleaned, or a broken front replaced without the trouble of cementing, the glass sides being sufficiently thin to allow the use of high power objectives.

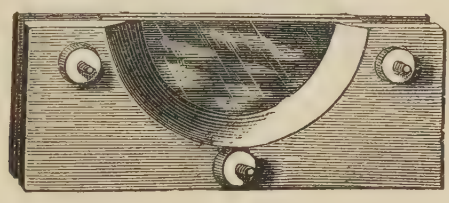


FIG. 75.

A microscopic life-slide also devised by Mr. Botterill is shown at Fig. 76; the advantages claimed for it are, the facility with which it can be used and cleaned; its reversibility, allowing either side of the object to be examined

FIG. 76.

through thin glass; the provision for renewing the supply of water without disturbing any part of the apparatus, thus enabling objects to be kept under examination for an indefinite period; the same arrangement also allowing of

the introduction of colouring matters, as carmine and indigo; and lastly, its moderate cost and durability.

For Confervæ, small Infusoria, and similar organisms it is sufficient to place the object on the bottom glass, with a drop of water, and apply the covering glass in same manner as when using a glass stage-plate. When a thicker layer of water is required, a narrow ring of vulcanite, cork, or other suitable material, of the requisite thickness, should be placed on the lower glass, and the object put in position, the covering glass being finally applied as in mounting objects in a cell. The supply of water can be maintained by putting a drop occasionally in one of the side "wells," keeping the slide, when not under examination, in a small damp chamber, to prevent evaporation. To change the water, supply through one "well," and draw out through the other by means of a roll of blotting paper.

Messrs. Thompson and Capper, of Liverpool, were the original makers of this slide, and also of Botterill's zoophyte trough, illustrated at Fig. 75.

The ordinary slip with a ledge of glass cemented to its lower edge, as shown in Fig. 77, is very useful, and saves the stage of the microscope from corrosion when marine organisms are being examined; it may easily be constructed by the student.

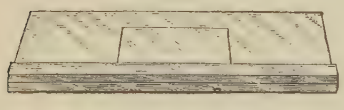


Fig. 77.

The glass for the construction of these troughs and slides may easily be cut with a glazier's diamond, and the edges ground parallel upon the flat face of an ordinary grindstone kept well wetted with water, or even by rubbing them upon a flat well-moistened piece of Yorkshire flagstone. An emery wheel will also answer the purpose. Pieces of superfluous glass may be removed by the use of a fine cut file lubricated with oil of turpentine, and holes made with

an ordinary drill lubricated with the same material. Glass may be turned to any shape in the lathe by using a smooth cut file, kept moistened with oil of turpentine, as the turning tool; but in order that this operation may be successful it is necessary that the piece for turning be not of too large diameter.

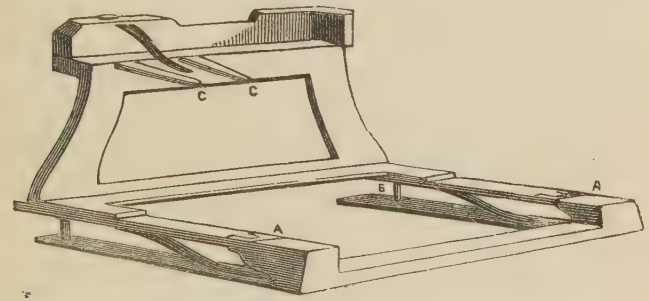


FIG. 78.

In working with high powers and expensive slides there is often a risk of either one or the other getting damaged, and this is especially the case with immersion objectives, where the kindly help of the dust on the cover-glass is not obtainable. It is never advisable to take high powers or rare slides to conversazione or other public meetings, on account of the miscellaneous character of observers; but if such a proceeding is imperative, the exhibitor should certainly provide himself with one of Stephenson's new safety stages, shown in Figs. 78 and 79, which may be the means of saving him a few regrets. It is so constructed that if by chance the object-glass is racked down on the thin cover,

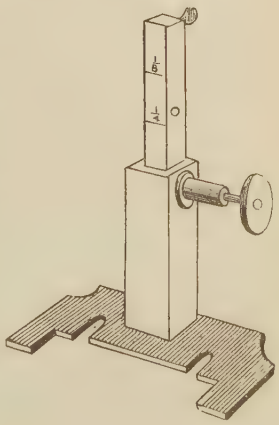


FIG. 79.

no damage is done, on account of the object receding as soon as contact is made, the springs shown in the figure making that motion possible. The object is placed on the two short arms C C, and is held in its place by the spring, which is placed above and between them. In order to make safety doubly sure Mr. Stephenson has devised a second piece of apparatus to act with the former; it is shown in Fig. 79, and consists of a square rod of brass which

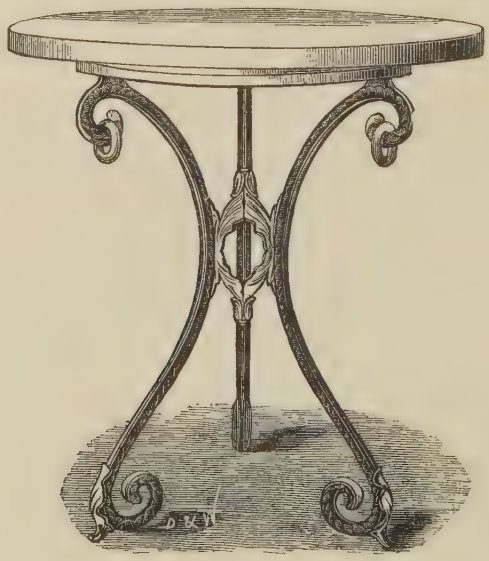


FIG. 80.

must be adjusted to suit the various objectives used; it is held in its place by a pin passing through it, attached to a screw at the outer side of the socket in which the rod slides. This instrument is placed (in the Ross model) beneath the bar which carries the microscope body, and, when properly adjusted, allows the objective to touch the object upon the stage, but arrests all further progress, no matter with what degree of force the coarse adjustment

may be pressed, a property of considerable value to public exhibitors.

Another very useful accessory is the revolving table, several forms of which are now sold at a very cheap rate. At one time the cheapest which could be obtained was about 9*l.*, and now they may be procured (with a slate top) for less than one-fourth of that sum.

When two or more microscopists are pursuing any investigation together, the constant rising from chairs must often have been thought a nuisance, but a cheap revolving table enables mutual observations to be made with comfort. The author's revolving table is 2 feet 4 inches in diameter, the top of it is 2 feet 3 inches from the ground, and four or even six observers may comfortably sit round it.

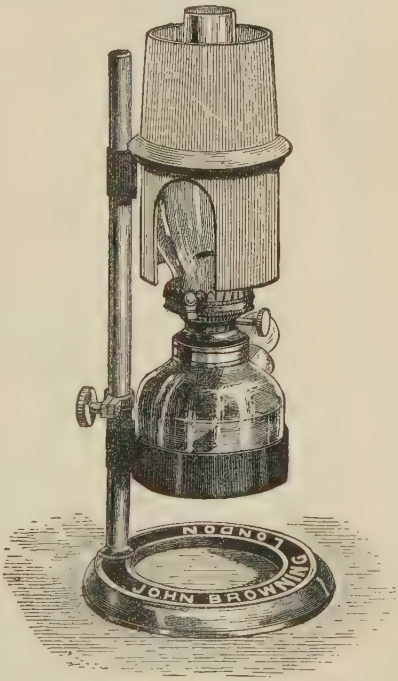


FIG. 81.

And now a few words as to illuminating apparatus. The best light which can be obtained is that from a good white cloud on a sunny day, but unfortunately in our towns and crowded cities we get but little sunlight undiluted with smoke, and students generally are occupied the day through, so that it becomes necessary to use artificial light.

When using light from the sky or from the sun, it should

be remembered that the rays are, for all practical purposes, parallel, and thereby differ essentially from artificial light, the rays of which converge strongly from the luminous centre. For use at home there is nothing, perhaps, so convenient as an argand gas reading lamp, sliding up and down on a metal rod, with a shade over it to prevent extraneous light from reaching the eyes. Students are very apt to work with too much light, and thereby impair the sensitiveness of their eyes; they should endeavour, however, to work with only just as much light as is necessary to bring out plainly the details of the object under examination, *and no more.*

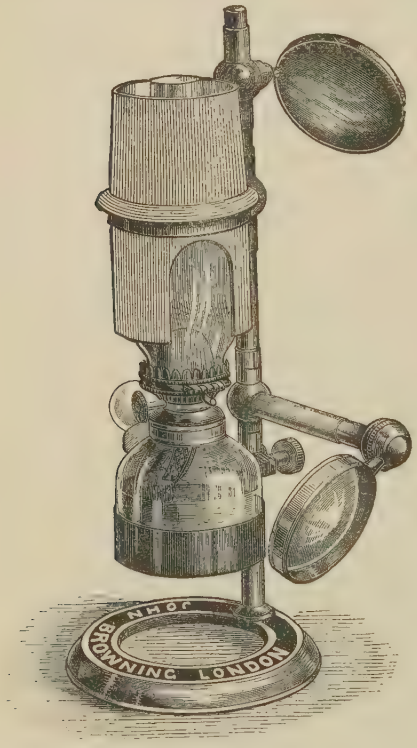


FIG. 82.

If an oil lamp is desired, a very common one may be made to answer almost every ordinary purpose, provided it is low enough, as when it is required to be raised, that may be readily accomplished by means of blocks of wood of varying thickness. The ordinary form of microscope lamp is shown in Fig. 81; it differs slightly in construction in the hands of different makers, but the student should eschew all forms in which the oil reservoir case is

soldered to the sliding ring; all the author has seen from different makers have come to pieces in a very short time. Browning's oil lamp, with bull's-eye condenser and silvered reflector, is shown at Fig. 82. It has a porcelain shade covering the chimney to protect the eyes from the excessive glare. This lamp (and indeed most oil lamps) is used with paraffin oil, and the brilliancy of the light may be increased by dissolving a little camphor in it. The light is more intense when the *edge* of the flame is turned towards the object to be illuminated; but if quantity of light is required rather than intensity, the flat side of the flame may be so disposed.

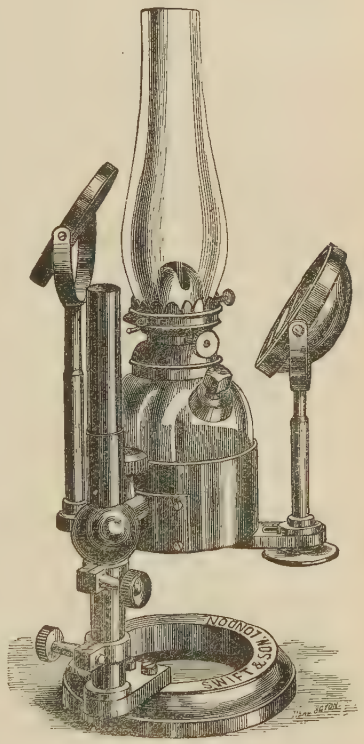


FIG. 83.

A rather better lamp than the above for general work is that of Mr. Swift, and shown in Fig. 83, which, however, in the writer's estimation, would be much improved by a glazed porcelain chimney with two opposite perforations, for in most examinations (perhaps all) it is a very important point to avoid a flood of extraneous light passing to the eyes.

There are several other lamps which may be mentioned here: Collins's Bockett lamp and Fiddian's lamp, made by Messrs. Ross and Co. The Fiddian lamp is supported by a massive claw stand, from which rises a vertical support on a

ball-and-socket joint. A brass tube slides on the vertical rod bearing the condenser and lamp with neutral tint shade and "white cloud" reflector having telescope and clamping screw adjustments. When these are placed in any desired relation to each other, the whole can be vertically adjusted by a rack and pinion with the greatest accuracy. Its price is five guineas. Another beautiful lamp has been made by Messrs. Wood, of Liverpool, for Messrs. Dallinger and Drysdale. These two observers, whilst working at the life-history of the monads, appreciated the difficulty of accurately centering the image of the flame when working with the $\frac{1}{25}$ and $\frac{1}{50}$ objectives, and so devised this lamp, which is illustrated in the April number of the 'Monthly Microscopical Journal' for 1876, vol. xv.

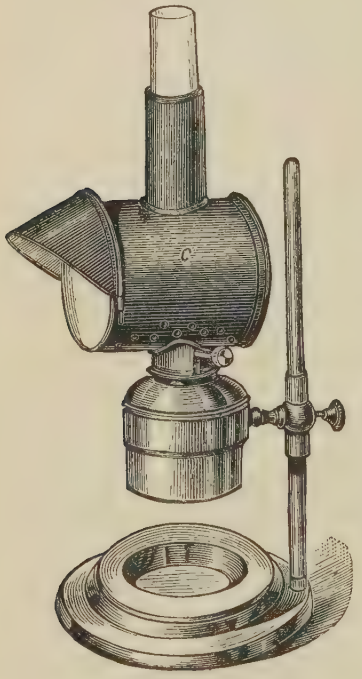


FIG. 84.

Parkes' microscope lamp with cooling evaporator may be seen in Figs. 84 and 85. C is a bronzed copper cylindrical shade $3\frac{1}{4}$ inches in diameter with a hood at the front to prevent the upward reflection of light. At the back is a parabolic reflector transmitting nearly parallel rays, made removable for the purpose of cleaning. At the front is a tinted "light-modifier," secured by a bayonet joint, and may be also removed when desirable. D is the "cooling evaporator"; a layer of thick felt is placed inside for saturation. When the lamp is

lighted, this vessel is filled with water, and so prevents the radiation of heat upon the observer's head. The felt requires moistening about once every five hours.

The light of the sun, a white cloud, or the electric light, which the author has used, and will illustrate in a future chapter, each give a light of remarkable purity. This is not the case, however, with the light from gas or from oil lamps. These last, especially gas sources of illumination, give a very objectionable yellow tone, while some tints are nearly suppressed. This effect has been noticed by all observers, and in 1872 Mr. Collins produced a light-corrector, and exhibited the same at a soirée at the Quekett Club. It consists of a brass stage-plate with a groove in which rotates a diaphragm of 4 apertures—one open, one fitted with a finely ground glass, while the others are fitted with two different tints of blue. Rainey produced a light-modifier before this, but it was of such construction that it required fitting to each microscope; that of Mr. Collins, on the other hand, can be used with any instrument, and without fitting.

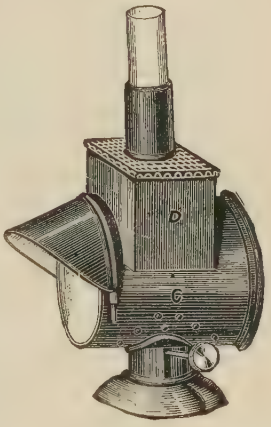


FIG. 85.

The effect of the blue glass is to effectually correct the yellowness proceeding from all artificial illumination, rendering the light soft and agreeable, as well as to improve the definition. To produce this effect, the writer uses a simple 3 in. × 1 in. slide of blue glass, such as is used by the chemist for the qualitative analysis of potash salts. It was obtained from Messrs. J. J. Griffin and Sons, Garrick Street, Covent Garden.

Dr. Woodward prefers to use the ammonia-sulphate of copper cell, and then only for high power definition, and he says he has been able to resolve the markings on *Amphipleura pellucida* with objectives found incapable of doing it with white light. Professor Smith, of Ashtabula, also expresses his approval of the use of monochromatic light. He says that with its use, and an eighth dry objective, he has easily resolved the *A. pellucida* to beads, in balsam, with deep eye-pieces; and with the lowest eye-piece the transverse and longitudinal striæ were easily seen.

The white cloud illuminator is a contrivance made in order to produce the same kind of illumination from artificial light as is obtainable from a white cloud. It is generally used with low powers only, and is made in several ways—a concave surface of plaster of Paris, a mirror coated at the back with zinc-white paint, roughened enamel, and white paper have all been used to produce this effect, as well as the disc of ground glass found in Mr. Collins's light-modifier.

Thus closes the chapter on accessories; but the student must not think we have exhausted the subject: there are many pieces of apparatus in occasional use which it has not been thought necessary to include here, and many others will be described in the subsequent chapters under the headings in which they are more intimately concerned.

CHAPTER V.

GENERAL REMARKS UPON OBJECTIVES—TEST OBJECTS.

WHEN we consider the many adjustments of apparatus needed ere a correct picture of an object can be placed before the eye, it will be readily seen how necessary it is to pay strict attention to details—more especially of illumination, this being one of the first and most important lessons the microscopist has to learn.

When rays of light pass through media with parallel faces, such as the glass slips used by every worker with the microscope, the emerging rays are parallel with those entering, the intermediate portion being bent away from both these planes as shown in Fig. 86. If water be used above the glass, the emergent ray will be bent up more towards the perpendicular, while when cedar-wood oil, or any of the homogeneous-immersion fluids are employed, the path of the ray will be one continuous line from the under side of the glass slip. A diagram of the passage of a light-ray through glass is shown in Fig. 86.

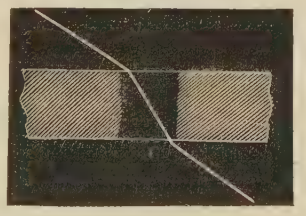


FIG. 86.

Ordinary glass slides, and also the thin covers, are made from crown glass having a refractive index varying from 1·5 to 1·525 referred to air as unity: the following table of mean refractive indices of many substances used by the microscopist may not be uninteresting:—

REFRACTIVE INDICES.

Air	1·000
Water	1·336
Sea water	1·343
Alcohol	1·373
Glacial acetic acid	1·380
Equal parts, glycerine and water	1·400
Glycerine	1·475
Oil of turpentine	1·478
Crown glass	1·5 to 1·525
Homogeneous-immersion fluid	1·500
Chloride of cadmium in glycerine	1·500
Cedar-wood oil	1·512
Canada balsam	1·532
Flint glass	1·575
Monobromide of naphthaline	1·658
Bisulphide of carbon	1·678
Oil of anise	1·811
Sulphur	2·115
Phosphorus	2·224

Plane or flat mirrors reflect an image of the same size as the object, the flame of a gas or oil lamp for instance, and therefore the rays are parallel, and the image is not inverted. When light is reflected from glass, the under side of which is silvered, much of it is lost from several causes; but when polished metal is employed for the reflecting surface, the rays do not enter the substance of the reflector, and there is less loss of light than in the former instance.

We must now turn our attention to the concave mirror, with which all respectable microscope stands are furnished. In this kind, the focus is situated at a point at which the reflected rays meet, and when rays parallel to the axis are brought together after reflection, the meeting point or focus is at an equal distance between the centre of curvature C and the mirror itself, and, consequently, if a luminous object be placed in this principal focus F, the rays emitted by the whole surface of the mirror will be parallel, as seen in Fig. 87.

If, however, the luminous point be at a greater distance

from the mirror than the principal focus, or *vice versâ* if the luminous rays fall divergent upon the concave mirror, a focus is obtained at another point called the conjugate focus. In the first case the rays, instead of being parallel, will converge towards L (Fig. 88), while in the second the focal

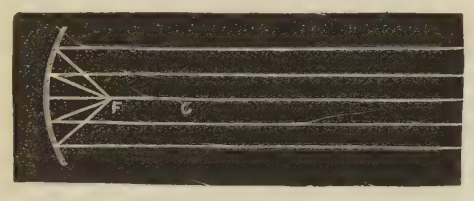


FIG. 87.

point of light will be removed further away from the mirror, and the rays proceeding from a lamp may be brought to a focus until the distance between the source of illumination and the mirror has been lessened to the centre of curvature, the rays being then reflected on to themselves.

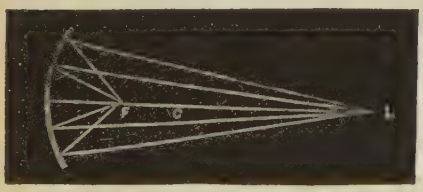


FIG. 88.

FIG. 89.

If the source of light be placed between the principal focus and the mirror the reflected rays will be divergent, as shown in Fig. 89.

Let us now consider the action of lenses upon illuminating rays. In a double convex lens the refracted rays from a parallel pencil of light form a focus very near to the centre of curvature of the lens, and conversely when a lamp is placed in its principal focus a double convex lens may be made to appear the source of light, as shown in Fig. 90.

It will be seen that a double convex lens has a principal focus on either side of it, and therefore the light may be parallelised on either side, but if the source of illumination be placed further away than the principal focus, the rays

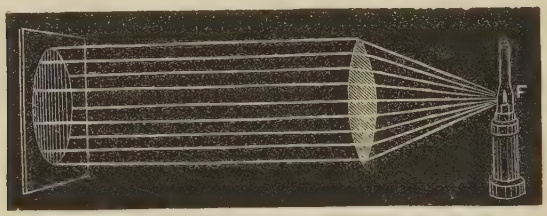


FIG. 90.

will be no longer parallel, but centred in a point at some distance from the opposite side of the lens, as shown in Fig. 91.

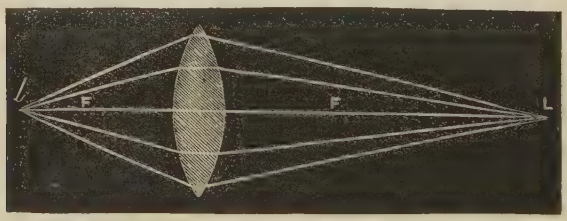


FIG. 91.

These points (*l* and L) are called conjugate foci, and do not lie in any fixed plane, but are dependent the one upon the other; it is this movement of the conjugate foci which yields a long working distance from the objective when the body of the microscope is shortened, and requires the object-glass to be approached nearer to the object when the draw-tube is used.

Diverging rays can be produced by placing the illuminating point between the principal focus and the lens, and

when converging rays fall upon a double-convex lens they are brought to a focus at a point between the principal focus and the lens itself, as shown in Fig. 92.

The action of a plano-convex lens, of which our bull's-eye condenser, Fig. 36, is a type, may be studied in the same diagrammatic manner. This may be considered as a double-convex lens split down the centre, and so forming two plano-convex lenses; it is generally used for purposes of microscopy on account of the great working distance of its focus. As already described, parallel rays falling upon a double-convex lens come to a focus very near the centre (radius) of its curvature, but when the same rays fall upon the curved surface of a bull's-eye condenser they are brought to a focus at a distance equal to the *diameter* of the curvature, or twice the distance of a double-convex lens, as may be seen in Fig. 93; and conversely if we wish to produce rays of parallel light from

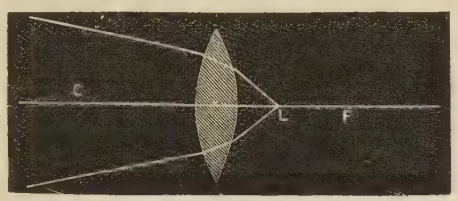


FIG. 92.

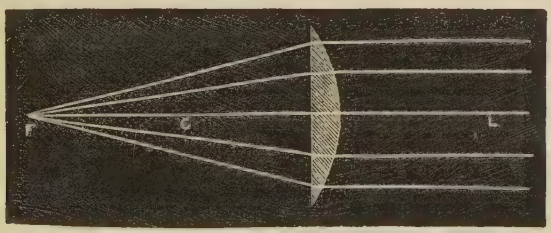


FIG. 93.

a lamp, the ordinary bull's-eye condenser must be placed twice as far from the luminous point as would be necessary in the case of a double-convex lens, a condition of

extreme importance when we consider the heat given out during combustion in most oil lamps.

These remarks upon the behaviour of certain lenses and mirrors towards the rays of light may be considered superfluous, nevertheless, as the student proceeds, he will find that not one word too much has been written. False appearances are often produced by the bad employment of light, and the student is advised to practise many kinds of illumination upon objects with which he may be familiar, so as to acquaint himself with the various appearances which diverse applications of it will afford.

When the action of the various mirrors and lenses has been fairly grasped, the student should proceed with some work capable of giving him experience of the manner in which various objects are delineated or depicted under various objectives. In the days of Dr. Goring (1832) when objectives even of high amplification had not surpassed an air angle of 55°, and even when achromatics were despised by nearly all working microscopists, it would not have been a difficult task to test an objective for its spherical and chromatic aberrations, by means of the tests we now possess; but time has changed all things microscopical; really bad lenses are rarities, and taking objectives to, and including the $\frac{1}{4}$-inch, it is remarkable how few imperfections they possess.

In order to correct perfectly the aberrations of objectives, the practical optician employs the globule of mercury or "artificial star" as a test object, while the accuracy of their setting is examined by studying the reflected image of a flame or the window bars, while the mount with lenses *in situ* is revolving in the lathe.

Beyond mention of it, the "artificial star" test need not be described here; it has been fully treated upon by Dr. Goring in the 'Microscopic Cabinet,' to which the

reader is referred, while the introduction of a good series of test objects renders the general employment of the former scarcely necessary.

It must not be forgotten that great differences exist between test objects. Amongst the slides of *Pleurosigma angulatum*, sold for this purpose, some are so extremely coarse as not to be a test in any sense of the word, while others are so finely marked that they can only be resolved with the greatest difficulty under a $\frac{1}{4}$-inch objective of high air angle. This is also the case with the diatoms *Navicula rhomboides* and *Amphipleura pellucida*, used as tests for the highest powers; so that no reliance should be placed upon statements that such and such a diatom was resolved under a certain objective.

Object-glasses for use with the microscope are usually spoken of as possessing the following qualities:—

1. Working distance of the front lens from the object;
2. Defining power;
3. Flatness of field and freedom from distortion;
4. Penetrating power;
5. Resolving power;

and it is to ascertain their excellence, or otherwise, in these directions that test objects are brought into use.

WORKING DISTANCE.—It has already been shown that the nomenclature of objectives does not presuppose any working distance from the front lens; in fact, such a thing would be impossible, seeing that the enlargement of the aperture reduces the distance of the front lens from the object, while the amplification remains the same. Their designation, such as a "1-inch objective," indicates only that such an object-glass should possess the same magnifying power as a single lens of 1-inch focus, the distance the

front of the system focusses from the object not being considered at all.

Gundlach, in a rather abstruse article in the 'American Monthly Microscopical Journal,' ii., 1881, tells us that "working distance" depends upon (1) the focal distance (nominal, it is presumed), (2) the aperture, (3) the number of lenses of which the objective is composed, (4) the proportionate curves of the lenses, and (5) the thickness of the lenses.

Great working distance is valuable in an objective only when circumstances demand it. Thus, for dissecting, or for the examination of opaque objects, a certain amount of distance is requisite for manipulation and illumination; but when an object has been prepared and mounted, no more working distance is absolutely required than will admit of the use of the thickest covering-glass, and of the examination of a moderate depth of object.

High-angle objectives of low power and consequent shorter working distance will define much better than the smaller apertures, and there is sufficient working distance, even for dissecting, with the 1-inch of 25° air angle.

There is some difficulty in selecting a $\frac{1}{2}$-inch objective. A glass of 40° air angle possesses considerable working distance, being a power well suited for dissections, used in conjunction with the erector; while a $\frac{1}{2}$-inch objective of 80° air-angle scarcely gives 0·03 inch of working distance, focussing closer to the object than an ordinary $\frac{1}{4}$-inch objective of 85°.

With the $\frac{1}{4}$-inch and all higher powers the working distance is very small, so that often the microscopist is precluded from using covering-glasses of the ordinary thickness. This is the case with all extremely high apertures used as dry objectives. Immersion objectives, for the same degrees of amplification, afford much longer

working distances than dry lenses, so that it is often possible to use an immersion $\frac{1}{16}$-inch where the covering-glass is too thick for a dry $\frac{1}{8}$th.

The author would like to see objectives catalogued by the makers in somewhat the following manner; the oculars too might be included:—

Designation.	Aperture.		At 10 inches with A ocular.		Price.	Remarks.
	Numerical.	Air angle.	Working distance.	Amplifying power.		
1-inch	..	16°	0·750	50	..	Triplet.
,, ...	..	25°	0·420	56	..	Dry objective.
$\frac{1}{2}$-inch	..	40°	0·300	100	..	,, ,,
,, ...	0·50	60°	0·150	106	..	,, ,,
,, ...	0·64	80°	0·032	110	..	,, ,,
$\frac{1}{4}$-inch	0·68	85°	0·080	200	..	,, ,,
,, ...	0·94	140°	0·038	210	..	Water-immersion.
$\frac{1}{12}$-inch	1·00	180°	0·024	600	..	Homogeneous-immersion.
,, ,,	1·43	..	0·007	615	..	
Oculars	A or 1	B or 2	C or 3	D or 4	E or 5	F or 6
Amplifying power in diameters	5·0	7·5	10·0	20·0	30·0	40·0

DEFINING POWER.—This property is of the first importance in objectives, and has been described by Dr. Goring to mean "nothing more than a destitution of both kinds of

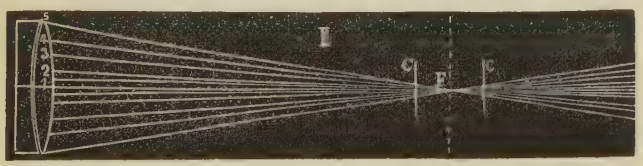


FIG. 94.

aberration." A well-corrected objective focusses the individual rays of a pencil of light, both from the centre and periphery, to the same plane as shown at F in Fig. 94; but, nevertheless, a more or less distinct image is produced for

some distance from each side of this focal plane. If, however, the rays are not thus corrected, the outlines of edges of the image will be thick and confused, and the glass is said "to be wanting in definition."

This fault may be shown diagrammatically in Fig. 95, which is an under-corrected glass, the peripheral rays being

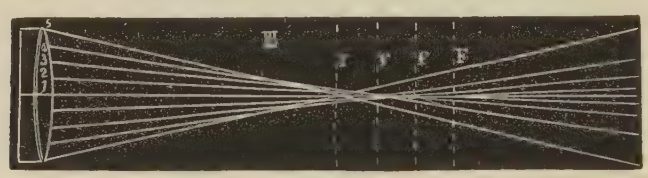


FIG. 95.

brought to a focus at *fff*, between the central focus F and the lens itself. Lenses in their primitive state are very much "under-corrected," and can only be employed when it is possible to cut off the peripheral rays by a diaphragm.

An over-corrected lens is shown in Fig. 96, from which it may be seen that the marginal rays are thrown further away from the glass, being brought to a focus at *fff* respectively, while F represents the focus of the central portion.

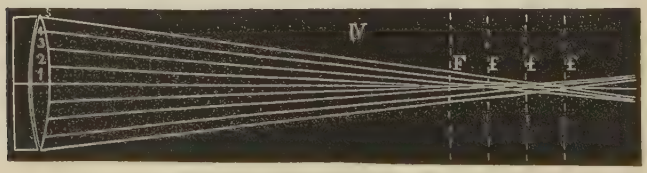


FIG. 96.

An objective free from both these defects is said to be free from spherical aberration—or aplanatic.

These defects in cheap low-angle objectives are corrected (if the term will apply) by the interposition of a diaphragm behind the back lens cutting off the marginal rays. These rays cannot then enter into the formation of the picture, the result being a dark, not very well defining glass of

low angle, but long working distance, though not sufficiently corrected to work with deep eye-pieces. In the best objectives of high angle the marginal rays are not cut off, but corrected to the very edges by the application of a wider back lens than usual; the aperture is therefore larger, but the objective possesses less penetration than one of lower aperture, and the working distance has been materially reduced.

A small aperture objective may be constructed from one of these more perfectly corrected lenses by the addition of a diaphragm; such an objective would bear deep eye-pieces and possess a fair amount of penetration, though the working distance may not be sensibly increased, owing to the thickness of the lenses and their various curves not being specially planned with this end in view.

The defining power of an objective may be examined by the employment of certain test objects obtainable from Messrs. Norman or Wheeler, until the student has learned how to prepare them for himself. The pollen of the Hollyhock (*Althea rosæ*), shown in Fig. 97, is a useful test; it must be illuminated as an opaque object, and with a deep eye-piece (the Huyghenian D), the minute spines should be readily and clearly defined.

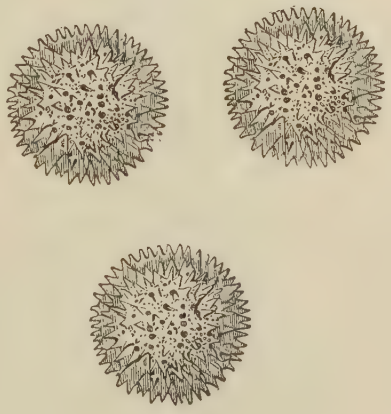


FIG. 97.

A well cut wood-section, such as is shown in Fig. 98, is also an excellent test of definition, the borders of each

vessel and cell should be clearly and sharply delineated, there must be no mistiness or blackness of edge. A dark

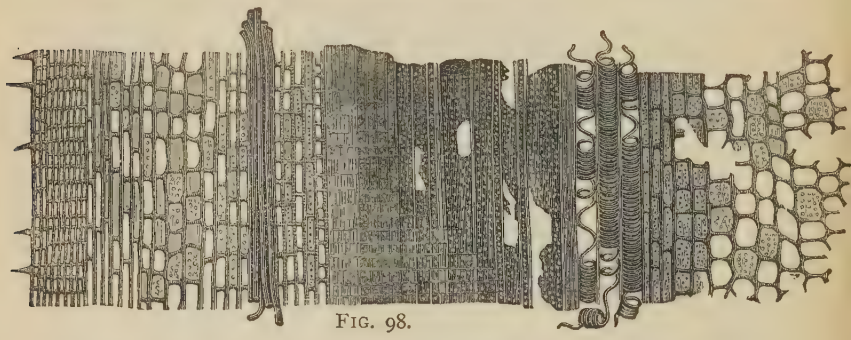


FIG. 98.

image shows at once that too much of the peripheral pencil has been cut off, the definition of a small aperture objective being *never* equal to one of large angle. The figure shows a section of the Horse-chestnut stem (*Æsculus hippocastanum*).

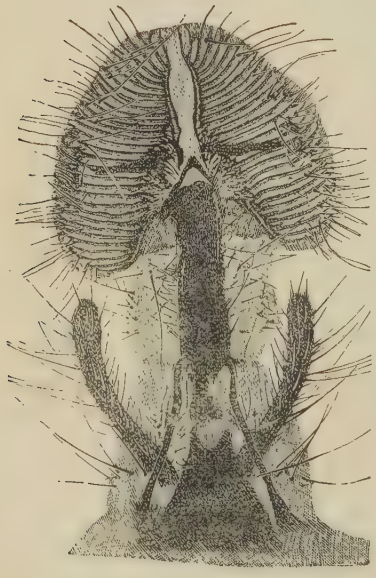


FIG. 99.

Triplets, such as are shown in Fig. 28, generally break down under deep eye-pieces.

The proboscis of the Blow-fly, shown in Fig. 99 —a favourite preparation of the late Mr. Topping —is an excellent object for testing the defining power of an objective of low amplification; the outlines may be fairly sharp, yet the details of the pseudo-tracheæ will not be clear, well defined, and free from colour under

a deep eye-piece, unless the corrections have been well cared for.

The student should, if possible, compare this object under two objectives, one of foreign make yielding an amplification of 50 diameters with the A eye-piece, the other a 1-inch of English construction, possessing an air angle of 30°.

Another exceedingly good test of definition is a well mounted specimen of the tracheal system of *Dytiscus marginalis*, delineated in Fig. 100.

In this instance the spiral threads should be visible without any halo of colour, and clearly separated so as to appear a continuous fibre rolled between two membranous walls.

The engraving shows the illusive appearance generally perceived in this object—that of watered silk — produced by the contact of the back and front of the spiral, at different inclinations, when the object is flattened and mounted in the usual way in balsam.

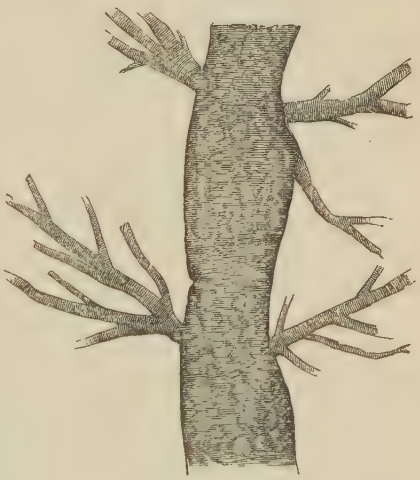


FIG. 100.

In order to discover how the correction for colour has been performed, several objects may be employed; Dr. Carpenter's test is the section of pine-wood shown in Fig. 101; it should be mounted dry, and the small circles (*glandulæ*) must be well defined and free from colour even with the D eye-piece. Perhaps *absolute* freedom from colour under deep eye-pieces does not yield the

utmost perfection in resolution; but authorities differ on this point.

For higher powers a white petal of the Pelargonium may serve as a test object, it is a moderately severe test for a

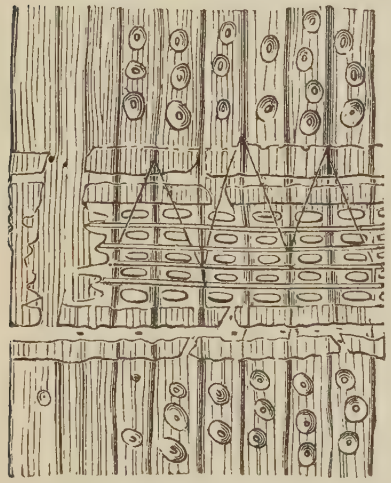


FIG. 101.

FIG. 102.

$\frac{1}{2}$-inch; while for higher powers still, the diatom *Meridion circulare* will show directly whether the corrections have been well executed.

Insect scales are generally used for judging the defining power of higher objectives, such as the scale of the *Morpho Menelaus* shown in Fig. 102, and that of the Podura, Fig. 103.

The former exhibits lines in a longitudinal direction with transverse markings, attributed to the corrugation of the internal surfaces of the lining membrane, and which are only to be noticed by the use of a good objective.

The Podura scale forms an excellent test of definition; it was known for this purpose before the appearance of

Pritchard's 'Microscopic Cabinet,' published in 1832; but the true character of the markings was not then known. This scale was then included amongst the "line tests," whilst now, with objectives of wide aperture, it presents

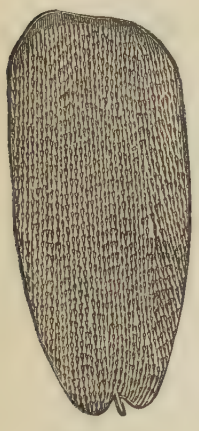


FIG. 103.

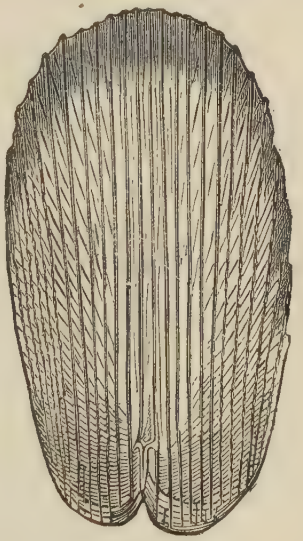


FIG. 104.

the appearance shown in the figure. Central light from an achromatic condenser is the best for exhibiting this scale, or it may be illuminated as an opaque object by means of the lamp and bull's-eye condenser.

Other insect scales are to be found in use as test-objects, such as those of the *Lepisma saccharina* shown in Fig. 104, and the battledoor scales of the *Polyommatus argus* delineated by Fig. 105.

These are considered much easier tests than those of the *M. Menelaus* and Podura (*Lepidocyrtus curvicollis*), but not so variable in quality. Fig. 106 shows one of the ordinary scales of the *Morpho Menelaus*, being, however, magnified to but one-half the extent of Fig. 105.

In the use of these test objects, great attention must be paid to the illumination and more particularly to the adjustment of the lenses for the thickness of the covering glass. But little correction is needed with the screw collar

FIG. 105.

FIG. 106.

adjustment in the water-immersion objective, and still less, if any, is required with the homogeneous system; nevertheless, it should always be added, as then correction is possible, should it ever be required to meet exceptional cases. In the lenses made by Messrs. Ross and Co., all the powers from the $\frac{1}{5}$-inch upwards can be used either as "dry" or "water-immersion," by merely moving the lenses by means of the adjusting collar near to the mark "wet," thus avoiding the cost of extra fronts and the inconvenience in changing them. Great care is required in adjusting them exactly, so as to get the best performance.

3. FLATNESS OF FIELD AND FREEDOM FROM DISTORTION.—These properties in objectives may be tested in several ways; for low powers, a section of a large spine of Echinus, such as shown in Fig. 107, may be used; a well cut and perfectly flat wood-section; or perhaps better still, one of Mr. Dancer's exquisite micro-photographs. The whole field should be well defined under

one focussing, the margin as well as the centre. For higher powers similar objects may be used, but of course of smaller dimensions, while freedom from distortion can

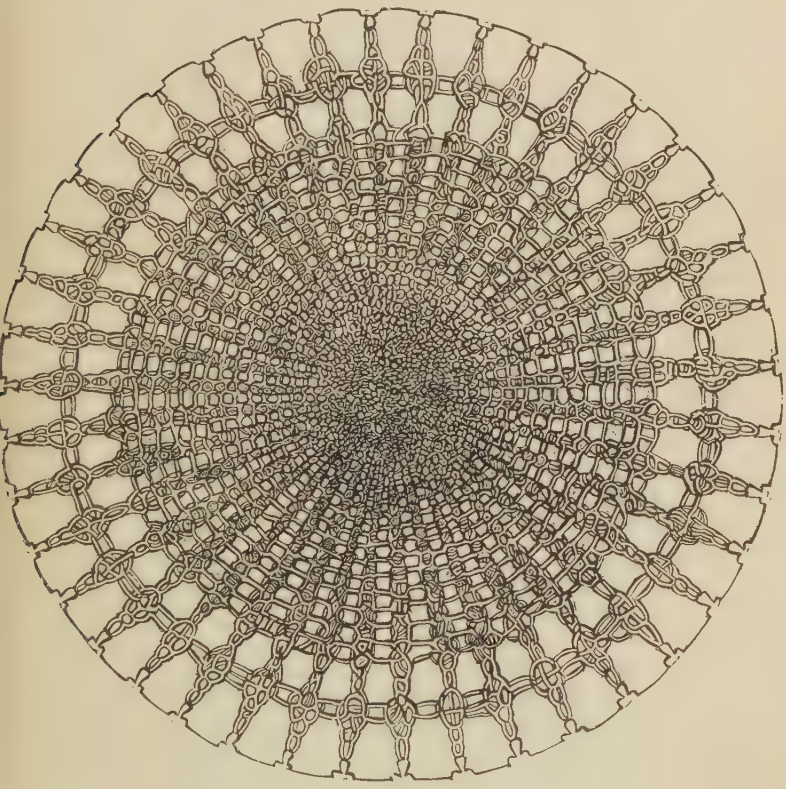


FIG. 107.

be tested for, by observation of the micrometer placed upon the stage, or a series of lines ruled by Wheeler, varying from one thousand to ten thousand to the inch.

4. PENETRATING POWER. — "Penetration," as it is understood now, signifies that property which an objective possesses whereby several planes of an object are

brought into focus simultaneously. Until very recently the property of *penetration* was shrouded in mystery and its usefulness often exaggerated, but thanks to Professor Abbe, who has made it a special study, most of the difficulties surrounding the subject have now been cleared away.

The greatest use of "penetration" is perhaps in the employment of the binocular microscope, for it is only when an object can be seen in its entirety that a true stereoscopic image can be obtained. There is no doubt also that this penetrating power is very useful for general work, such as dissecting; still it should be remembered that a low-angle lens with much penetration will not usually stand deep eye-pieces. Dr. Blackham writes of "depth of focus" as "only a residual error at best" and "that it can be left in a wide-angle lens as well as in a narrow-angled one." Whether this is practically correct, perhaps opticians will be able to tell us, but it seems to the author much more difficult to leave spherical aberrations uncorrected and practically unnoticed in a wide-angle lens than in one of narrow angle.

Professor Abbe has demonstrated the fact that penetration or "depth of vision" depends upon two circumstances: first, the *accommodation depth* of the eye, and secondly the *depth of focus*, varying with the quality of the images produced from certain distances on each side of the exact focal plane. The eye, being insensible to small defects in the various images, can practically discern the object in its several depths, provided the images compare with sufficient exactness.

The accommodation depth is very great with low amplifications. Under a magnifying power of 10 diameters it amounts to nearly 2·1 millimetres, while with higher powers the image passes quickly into a mere transverse

section. The depth of focus does not diminish at such a rapid rate, but the thickness of an object which can be seen under one focussing decreases accordingly as the amplification increases, and therefore it is most important, where binocular vision is essayed, to use the lowest power sufficient for distinctly recognising the object; and with transmitted light to employ as narrow a pencil as will sufficiently illuminate it.

The following table by Professor Abbe will sufficiently illustrate these remarks:—

Amplification. / 0·50 N.A.	*a*. Diameter of Field.	*b*. Accommodation Depth.	*c*. Focal Depth.	*d*. Depth of Vision. / $b + c$.	Ratio a to d. / $\frac{a}{d}$
	mm.	mm.	mm.	mm.	
10	25·0	2·08	0·073	2·153	11·6 to 1
30	8·3	0·23	0·024	0·254	32·7 ,,
100	2·5	0·02	0·0073	0·0273	91·6 ,,
300	0·83	0·0023	0·0024	0·0047	176·6 ,,
1000	0·25	0·00021	0·00073	0·00094	266 ,,
3000	0·083	0·00002	0·00024	0·00026	319 ,,

The higher the numerical aperture of an objective the less will the penetration be, though the defining and resolving power, with quantity of light admitted, increase with the absolute aperture of the objective, provided the aberrations are well corrected.

The following table (see next page) has been abstracted from the 'Journal of the Royal Microscopical Society' for August 1881, wherein may also be found Professor Abbe's paper from which we have largely quoted.

From the foregoing considerations, "tests" for penetration would seem to be superfluous, seeing that it results from two almost fixed conditions; nevertheless, it is well to know how it may be observed. A section of frog's lung or of human liver (Fig. 108) is useful for this purpose, as when mounted, the various parts will be found to have con-

Numerical Aperture. ($n \sin u = a$.)	Penetrating Power. $\left(\frac{1}{a}\right)$	Illuminating Power. (a^2.)	Numerical Aperture. ($n \sin u = a$.)	Penetrating Power. $\left(\frac{1}{a}\right)$	Illuminating Power. (a^2.)
1·52	·658	2·310	1·00	1·000	1·000
1·50	·667	2·250	0·98	1·020	0·960
1·48	·676	2·190	0·96	1·042	0·922
1·46	·685	2·132	0·94	1·064	0·884
1·44	·694	2·074	0·92	1·087	0·846
1·42	·704	2·016	0·90	1·111	0·810
1·40	·714	1·960	0·88	1·136	0·774
1·38	·725	1·904	0·86	1·163	0·740
1·36	·735	1·850	0·84	1·190	0·706
1·34	·746	1·796	0·82	1·220	0·672
1·33	·752	1·770	0·80	1·250	0·640
1·32	·758	1·742	0·78	1·282	0·608
1·30	·769	1·690	0·76	1·316	0·578
1·28	·781	1·638	0·74	1·351	0·548
1·26	·794	1·588	0·72	1·389	0·518
1·24	·806	1·538	0·70	1·429	0·490
1·22	·820	1·488	0·68	1·471	0·462
1·20	·833	1·440	0·66	1·515	0·436
1·18	·847	1·392	0·64	1·562	0·410
1·16	·862	1·346	0·62	1·613	0·384
1·14	·877	1·300	0·60	1·667	0·360
1·12	·893	1·254	0·58	1·724	0·336
1·10	·909	1·210	0·56	1·786	0·314
1·08	·926	1·166	0·54	1·852	0·292
1·06	·943	1·124	0·52	1·923	0·270
1·04	·962	1·082	0·50	2·000	0·250
1·02	·980	1·040			

tracted somewhat, producing corrugations or folds, and consequently the rays proceed from different planes, so that the test should be to see how much or how little of the total depth can be seen under one focussing, without indistinctness.

As an instance of the work specially suited for low angles and consequent penetration, the cyclosis in *Vallisneria spiralis* is often cited. The author can assure his readers that this may be easily seen with advantage under a $\frac{1}{2}$-inch objective of 80° air angle (0·64 numerical aperture), the largest yet made for this power in this country.

5. Resolving Power.—Without entering into any of the theories of this property in objectives, it may be briefly

stated that it depends entirely upon large aperture, combined, of course, with accuracy of the corrections for sphericity and chromatism. The several pieces of apparatus mentioned in the previous chapter are often essential with objectives of wide aperture, for it is obvious that the advantages would be lost if sufficiently oblique rays did not enter into the formation of the image. The Ross-Zentmayer stand, used with a 1-inch or 2-inch objective as a condenser, the radial substage condenser of Messrs. Swift and Son, or the oil-immersion condenser of Messrs. Powell and Lealand, are all capable of producing light-rays of sufficient obliquity.

FIG. 108.

Diatom frustules, as a rule, furnish tests for the resolving property of medium and high-power objectives, to which may be added the insect scales already shown; but these natural tests are all of very variable quality. *Gyrosigma formosum* and *G. angulatum*, shown by Figs. 109 and 110 respectively, are tests of the resolving property of the $\frac{1}{2}$ and $\frac{1}{4}$-inch objectives, both of which have been engraved from photographs taken by the late Dr. Redmayne, of Bolton.

Navicula rhomboides, shown in Fig. 111, is used as a test for the $\frac{1}{8}$th objective and higher powers. It is rather a difficult diatom to resolve properly without accessories, but with the Powell and Lealand oil-immersion condenser it may be managed without much trouble.

Amphipleura pellucida, shown in Fig. 112, is a most difficult diatom to resolve; indeed, it cannot be accomplished by any dry objective save of the widest aperture, and even then it requires most careful attention to the details of illumination.

Dr. Woodward considers this frustule to be the most useful test for immersion objectives of $\frac{1}{8}$ power and higher. The resolution into lines is not so

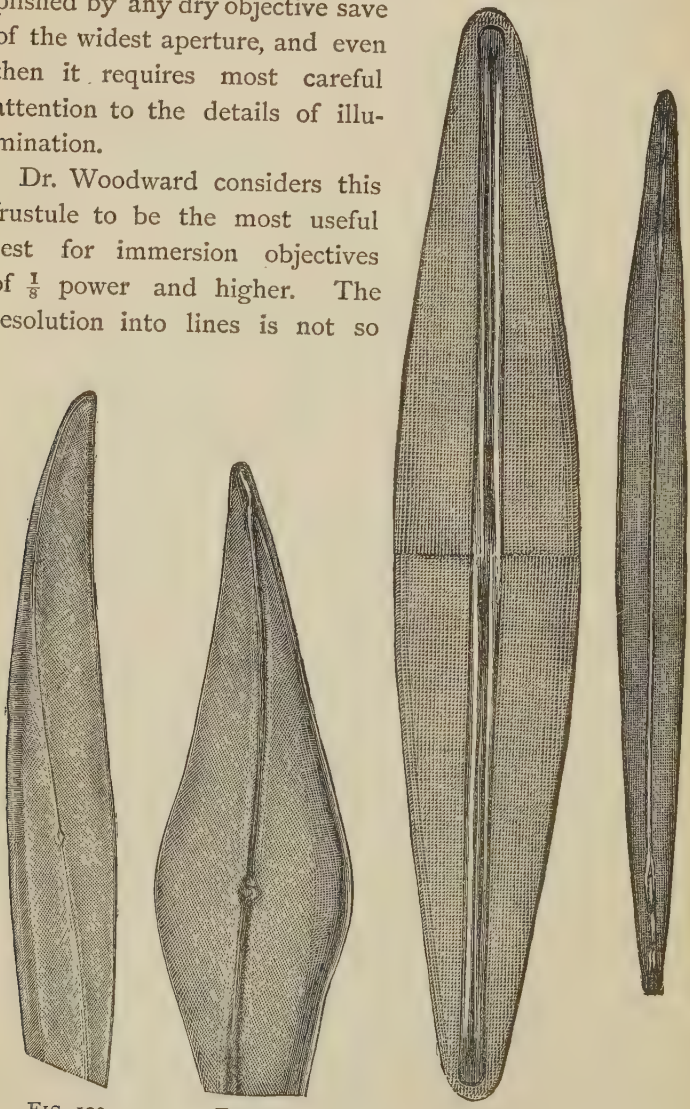


FIG. 109. FIG. 110. FIG. 111. FIG. 112.

difficult, when compared with that into dots. Mr. Wenham, when describing his new illuminator for diatoms, states that *he* was never successful in the patient manipulation required to resolve this diatom by the old methods of illumination.

Allusion has already been made to the variations likely to occur in the markings upon the frustules of diatoms and the scales of insects. In order to avoid these irregularities, the late Herr Nobert of Pomerania issued a series of test lines ruled upon glass, each band containing lines of a definite number to the inch. The most popular is that known as the 19-band plate, containing lines to the inch as under :—

Band.	Number of Spaces per Inch, about.	Band.	Number of Spaces per Inch, about.
I.	11,300	XI.	68,000
II.	17,000	XII.	73,000
III.	22,500	XIII.	79,000
IV.	28,000	XIV.	84,000
V.	34,000	XV.	90,000
VI.	39,800	XVI.	96,000
VII.	45,500	XVII.	101,000
VIII.	51,200	XVIII.	106,500
IX.	56,800	XIX.	112,000
X.	62,500		

Herr Nobert often expressed his opinion that the last four bands of this plate would never be resolved by any objective ; but after inspecting Dr. Woodward's photographs of the whole series, he produced another "plate" ruled to the twentieth band, the tenth on which corresponds to the nineteenth on the old, the twentieth band being ruled at the rate of 200,000 lines to the inch.

Herr Möller produces what is called a "test-platte," containing 20 diatoms, mounted dry or in balsam; they are arranged upon the slide in a row, at the beginning and end of which is a specimen of *Eupodiscus argus*. The

following is a list of the diatoms on this "test-platte," with the number of striations to the inch, as given by Morley :—

Diatom.	Number of Striæ to the Inch.
1. Triceratium favus	3,060 to 3,080
2. Pinnularia nobilis	10,800 ,, 12,500
3. Navicula lyra	16,300 ,, 18,500
4. ,, ,, ...	25,000 ,, 27,000
5. Pinnularia interrupta	26,500 ,, 26,800
6. Stauroneis Phœnicenteron	31,100 ,, 33,000
7. Grammatophora marina	36,300 ..
8. Pleurosigma Balticum	31,500 ,, 34,300
9. ,, acuminatum	42,700 ..
10. Nitzschia amphyoxis	42,900 ,, 45,300
11. Pleurosigma angulatum	43,800 ..
12. Grammatophora subtilissima	61,200 ,, 61,700
13. Surirella gemma	51,400 ,, 54,800
14. Nitzschia sigmoida	63,000 ,, 63,300
15. Pleurosigma fasciola	55,500 ,, 56,500
16. Surirella gemma	63,000 ,, 70,400
17. Cymatopleura elliptica	63,300 ..
18. Navicula crassinervis	79,400 ,, 82,200
19. Nitzschia curvula	84,500 ,, 84,700
20. Amphipleura pellucida	92,700 ,, 92,900

It has already been mentioned that the resolving power of objectives depends entirely upon their aperture, with the excellence of their corrections for colour and sphericity.

Objectives of low angle are generally made with posterior lenses of such a size as to exclude the extreme uncorrected marginal rays, or if this is not done the margins are cut off by a diaphragm. The utilised portion is very fairly corrected *for colour*, but nevertheless the spherical aberrations, though small, must exist ; on the other hand, wide apertures require large back lenses, the rays from which cannot be cut off by a diaphragm without reducing the angle, and therefore the corrections have to be applied to the more oblique pencils ; and chromatic aberration can scarcely be corrected without at the same time affecting the spherical in a very great degree.

The value of wide pencils was first put into practical form by Hartnack, who made in the ordinary course of business water-immersion objectives utilising pencils of light approaching to 170° in air; a drop of water was placed between the objective and the object, and thus by passing through a denser medium, rays of light entered the object-glass which could not possibly enter from air, giving at the same time a considerable working distance for the higher powers. Some years afterwards Mr. J. W. Stephenson, whose name has been already associated with the erecting binocular, conceived the idea of substituting for water, in the immersion objective, a fluid having the same refractive and dispersive power as crown glass; fourfold systems upon this plan were calculated by Professor Abbe and made by Carl Zeiss, the optician of Jena, of $\frac{1}{12}$ and $\frac{1}{8}$ equivalent foci. These objectives had a balsam angle of 113°: greater than the maximum of 180° in air, in the ratio of 5 to 4.

Messrs. Powell and Lealand have recently constructed a homogeneous-immersion objective of $\frac{1}{12}$-inch equivalent focus, with a numerical aperture of 1·43 or 140° balsam angle, with two extra fronts, one of which gives an aperture of 1·28 or 115° balsam angle, while the other provides an aperture of 1·0, or 82° in balsam. With an aperture of 1·43 the working distance is 0·007 inch; the aperture of 1·28 gives a focal distance of 0·016 inch; while with the numerical aperture of 1·0 the working distance is 0·024 inch.

It was the existence of these immersion objectives and their various angular values which led Professor Abbe to investigate the general principles of microscopic vision. He tells us that "the very first step of every understanding of the microscope is to abandon the gratuitous assumption of our ancestors, that microscopical vision is an *imitation* of

macroscopical, and to become familiar with the idea that it is a thing *sui generis*, in regard to which nothing can be legitimately inferred from the optical phenomena connected with bodies of large size."

Professor Abbe discovered the fact that the microscopical image is the result of diffraction, or the consequence of those changes which are produced in rays of light by their interception by minute particles; the rays are collected at the back of the objective, where they depict the direct and spectral images of the source of light, reaching in their further course the plane which is conjugate to the object, and give rise there to an interference phenomenon, which gives the ultimate image observed by the eye-piece, and, therefore, the image depends essentially on the number and distribution of the refracted beams which enter the objective.* From this it appears that the larger the number of diffracted rays admitted into the objective the greater likeness to the object will the image possess, a true image being only produced when *all* the diffracted rays from the object are admitted.

Dr. G. Blackham, in a paper read before the Microscopical Congress at Indianapolis, August 15, 1878, said, "Now, if it is the function of the objective to collect and bring to a focus rays of light too divergent to be received by the unaided eye . . . the more of these lost rays that a given glass can so collect and bring to a focus the better the glass," and "one would naturally expect to find that the improvement or evolution of the microscope was accompanied by an increase of the angular aperture of the objectives, and this, indeed, we find to be the case." Un-

* It is clearly beyond the scope of a work such as this to enter fully into the details of this most important question. Those who wish for a more complete dissertation are referred to the April number of the 'Journal of the Royal Microscopical Society' for 1881, where also Professor Abbe's paper "On the Estimation of Aperture" may be found.

aided, the human eye will only admit pencils of about 10°, and as light is dispersed from every point of an object to an angle of 180°, those glasses which approach infinitely near this latter angle must give a more correct image of the object that those of small apertures.

When we appreciate the above facts we shall be able to estimate the value of immersion objectives, which enable us to collect and gather to a focus rays which cannot possibly enter the microscope when a film of air exists between the objective and object. The editors of the 'Journal of the Royal Microscopical Society' have given diagrams of the relative diameters of the utilised back lenses of dry and immersion objectives of the same power, commencing with an air angle of 60° and ending with the homogeneous hemisphere of 180° balsam angle, which has nearly the same refractive index as crown glass. They also add, "Thus, if we commence with an air angle of 10° and proceed by successive additions of 10° up to 180° air angle, passing then to 82° balsam angle, and again progressing to the nearest practicable approximation to 180° balsam angle, the emergent pencils will show a continuous increase; there is no break at 180° air angle, nor does anything abnormal appear at that point, but we have a regularly progressive series from the lowest air angle to the highest balsam angle."

It has been urged by some, that all those rays equivalent to more than 180° air angle are not image-forming rays, but this assertion has also been dealt with in the same journal. "The simplest experiment of all is to take a homogeneous-immersion objective of large aperture, say 1·25 (110° balsam angle), and place a stop of tin-foil on the back lens, leaving only a small clear annulus of the extreme marginal rays. With sufficient obliquity of the illumination the image of the object will be seen perfectly delineated either on a bright or dark field."

An immense amount of discussion took place in reference to these lenses in England, nearly all the opponents to the system averring that by the use of an immersion fluid the aperture was *cut down;* that, in fact, rays which were equivalent to more than 180° in air were of no practical utility, even if they could be made to enter an object-glass and form an image. The editors of the 'Journal of the Royal Microscopical Society,' on page 305 of the 'Journal,'* write:—"The 'aperture question' will ever hold a most prominent place in the history of the microscope, representing as extraordinary a series of mistakes as were ever committed in any branch of science, and in which (down to comparatively recent times) both the leaders and the rank and file were equally involved. 'Aperture' may be said to have been the *haschische* of the microscopist; when *that* has formed the subject of consideration, the simplest and oldest established optical principles have not been disregarded merely, but their very converse tacitly assumed, as if the great optical physicists of this and the previous century had never lived or had written nothing that was worthy of consideration!"

Since penning the foregoing remarks, the author has been informed by Mr. J. B. Dancer, of Manchester, that for some years he has practically effected the reduction of aperture in somewhat a similar manner to that indicated on page 107. Instead, however, of screwing in a diaphragm of definite aperture behind the back lens of the objective, he uses an ingeniously constructed graduating diaphragm, screwing into the lower end of the microscope body as an adapter, into the lower end of which the objective is made to fit.

Mr. Dancer constructed this, some years ago, to give *penetration* to wide angle lenses, and has sent the author

* Series ii. vol. i. part 2.

the apparatus in question. There has scarcely been time to thoroughly test the apparatus while this sheet is passing through the press—the author has, however, tried it upon a $\frac{1}{2}$-inch objective of wide aperture, and this was cut down in such a manner as to enable it to be used with the Wenham binocular without producing any distortion.

The working distance of the front lens was not in the least altered by the contraction of the diaphragm, so that the objection to most wide-aperture lenses still remains, when distance from the object, either for illumination or for use of dissecting instruments, is required.

Having become somewhat confused in our nomenclature of apertures, on account of the unequal value of the same as expressed in degrees, Professor Abbe introduced a system based on his own experiments and used in harmony with existing but older optical laws. Under his hand the air angle of 180° was identical with the water angle of 97° and the balsam angle of 82°, but instead of giving them three separate values he introduced the term "Numerical Aperture," equivalent to 1·0 in each of the three above instances. The *numerical aperture* is easily obtained by multiplying the sine of the semi-angle by the refractive index of the fluid in which that angle has been measured: this is the meaning of the formula:

$$n \sin u = a,$$

where n = the refractive index of the medium; sin u = the sine of the semi-angle, while a = the numerical aperture.

A table of natural sines for each degree from 1° to 90° may be found in Chapter IX.

The following table has been printed for many months upon the cover of the 'Journal of the Royal Microscopical Society' (see next page).

Diameters of the Back Lenses of various Dry and Immersion Objectives of the same Power (⅛ in.) from 0·50 to 1·52 N. A.	Numerical Aperture. ($n \sin u = a$.)	Angles of Aperture ($= 2u$). Dry Objectives. ($n = 1$.)	Angles of Aperture ($= 2u$). Water-Immersion Objectives. ($n = 1{\cdot}33$).	Angles of Aperture ($= 2u$). Homogeneous Immersion Objectives. ($n = 1{\cdot}52$.)	Illuminating Power. (a^2.)	Theoretical Resolving Power, in Lines to an Inch. ($\lambda = 0{\cdot}5269\,\mu$ = line E.)
		° ′	° ′	° ′		
	1·52	..	..	180 0	2·31	146,528
	1·50	..	..	161 23	2·25	144,600
	1·48	..	..	153 39	2·16	142,672
	1·46	..	..	147 42	2·13	140,744
	1·44	..	..	142 40	2·07	138,816
	1·42	..	..	138 12	2·02	136,888
	1·40	..	..	134 10	1·96	134,960
	1·38	..	..	130 26	1·90	133,032
	1·36	..	..	126 57	1·85	131,104
	1·34	..	..	123 40	1·80	129,176
	1·33	..	180 0	122 6	1·77	128,212
	1·32	..	165 56	120 33	1·74	127,248
	1·30	..	155 38	117 34	1·69	125,320
	1·28	..	148 28	114 44	1·64	123,392
	1·26	..	142 39	111 59	1·59	121,464
	1·24	..	147 36	109 20	1·54	119,536
	1·22	..	133 4	106 45	1·49	117,608
	1·20	..	128 55	104 15	1·44	115,680
	1·18	..	125 3	101 50	1·39	113,752
	1·16	..	121 26	99 29	1·35	111,824
	1·14	..	118 00	97 11	1·30	109,896
	1·12	..	114 44	94 56	1·25	107,968
	1·10	..	111 36	92 43	1·21	106,040
	1·08	..	108 36	90 33	1·17	104,112
	1·06	..	105 42	88 26	1·12	102,184
	1·04	..	102 53	86 21	1·08	100,256
	1·02	..	100 10	84 18	1·04	98,328
	1·0	180 0	97 31	82 17	1·00	96,400
	0·98	157 2	94 56	80 17	·96	94,472
	0·96	147 29	92 24	78 20	·92	92,544
	0·94	140 6	89 56	76 24	·88	90,616
	0·92	133 51	87 32	74 30	·85	88,688
	0·90	128 19	85 10	72 36	·81	86,760
	0·88	123 17	82 51	70 44	·77	84,832
	0·86	118 38	80 34	68 54	·74	82,904
	0·84	114 17	78 20	67 6	·71	80,976
	0·82	110 10	76 8	65 18	·67	79,048
	0·80	106 16	73 58	63 31	·64	77,120
	0·78	102 31	71 49	61 45	·61	75,192
	0·76	98 56	69 42	60 0	·58	73,264
	0·74	95 28	67 36	58 16	·55	71,336
	0·72	92 6	65 32	56 32	·52	69,408
	0·70	88 51	63 31	54 50	·49	67,480
	0·68	85 41	61 30	53 9	·46	65,552
	0·66	82 36	59 30	51 28	·44	63,624
	0·64	79 35	57 31	49 48	·41	61,696
	0·62	76 38	55 34	48 9	·38	59,768
	0·60	73 44	53 38	46 30	·36	57,840
	0·58	70 54	51 42	44 51	·34	55,912
	0·56	68 6	49 48	43 14	·31	53,984
	0·54	65 22	47 54	41 37	·29	52,056
	0·52	62 40	46 2	40 0	·27	50,128
	0·50	60 0	44 10	38 24	·25	48,200

CHAPTER VI.

THE COLLECTION OF OBJECTS.

It has been the author's endeavour to persuade the student to take up some special branch of study with the aid of the microscope, and with this end in view the following chapter has been written, showing where certain objects are to be found, what apparatus is requisite for their collection, what to collect, how to collect, and when gathered, how to preserve them for future examination under the microscope.

Collectors of experience will not require to be informed on many of these points, and therefore to make the chapter interesting to more than the mere student a list of works treating on each subject has been appended, and also the names of several species under each heading in order that the possessor of a microscope may know what slides to purchase should he desire to fill a cabinet in that manner.

The author would strongly advise the young student to refrain from flitting hither and thither over the whole range of microscopical objects. It is not enough to be able to name a few rotifers or rare diatoms, such knowledge is of the shallowest kind ; but if he sets himself to work to study the life-history of some hitherto obscure organism, or the anatomy of an insect, the outward form of which he is alone familiar with, he may rely upon it, he will be useful in his generation.

Most collectors have their own method of gathering specimens, and are very conservative on this point, but the

telescopic walking-stick with all its fittings as shown in Fig. 113, is an article generally used by all. There are generally supplied with it, a ring to carry a fine muslin net, a ring to hold a bottle, a weed knife, a spoon, and a drag hook for weeds.

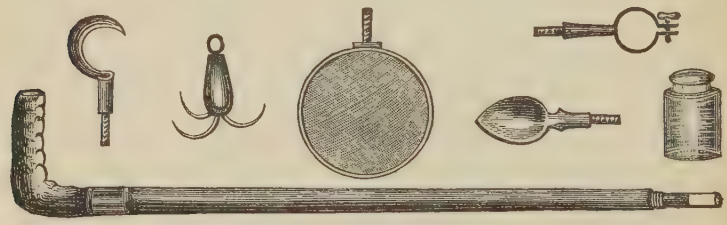


FIG. 113.

Mr. Baker, of Holborn, supplies the article in rather a different form: the bottom ring is not clamped by a screw as shown in the figure, but is furnished inside with a thread, into which is made to screw the neck of one of the York Glass Co.'s bottles.

For many purposes a pond scoop is required, such as for scraping the surface of the mud at the bottom of pools when searching for Oscillatoria, &c., and if it is made to screw into the end of the collecting stick it will be very convenient. It is simply a ring of tinned iron about 5-inches in diameter and 1-inch deep. Both edges are "wired," as the tinsmiths call it, so that a piece of thin muslin or stout gauze may be stretched tightly over it. It is shown in Fig. 114.

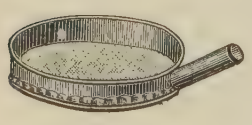


FIG. 114.

Some collectors prefer to secure the muslin over the scoop with a firm elastic band, so that after collecting it may be removed and folded up for transport home—and perhaps this is the better plan.

The tow-net, Fig. 115, is of great use in collecting marine, and even river or lake objects. It is made of fine but strong muslin, tied at the large end round a wooden hoop, while the nethermost extremity is secured round a small wide-mouthed bottle, so that the more delicate organisms may find their way into it, and so be out of the way of the currents caused by the passage of the net through the water.

Fig. 115.

The tow-net, as illustrated, is furnished with an interior net, which, acting as a valve, prevents the escape of organisms which have once been enclosed.

Aquatic organisms, whether animal or vegetable, are met with in all kinds of water; even the tap water supplied by some of our corporations is extremely rich in specimens, while in clean ponds the collector will not fail to find a host of treasures; in impure streams and pools, containing sewage and other decomposing matters, only such common animalcula as *Paramecium aurelia* are to be found. Other objects are fixed upon stones and weeds under water, and little pieces of dead stick are often found covered with interesting objects.

When the water is not rich in specimens, it may be necessary to concentrate them by straining off the superfluous water, which may be effected by using the filter shown in Fig. 116. It consists of two small funnels passing through a cork as shown in the figure, the one which is

inverted in the bottle being covered at the mouth with very fine muslin. The water containing the organisms is poured in at the top funnel, while water only issues from the stem of that inverted in the water.

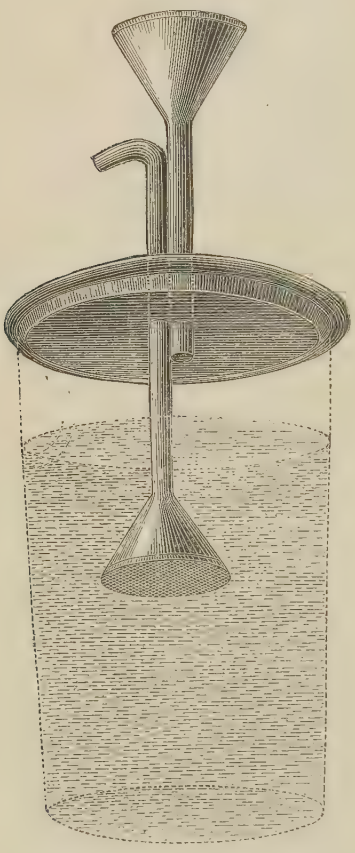


FIG. 116.

FIG. 117.

This operation may be continued until the bottle is well stocked, when the contents may be carried homewards for examination.

When an organism is required to be removed from a

bottle of water, a tube or tubes of the form shown in Fig. 117 will be found necessary. They may be cut from ordinary glass tubing by making a cut with the edge of a three-square file, and breaking it in two with the fingers. The sharp edges should then be fused by holding in the flame, finally allowing to cool gradually. The bent tubes may be made by taking a length sufficient for two tubes and softening the middle portion in the flame of an ordinary gas burner or spirit lamp, and when sufficiently softened the two extremities are to be pulled asunder so as to form a couple of tubes of the form of B, Fig. 117, they can then be cut asunder with the file and the edges fused. The form A is produced in a similar manner, the softened portion being drawn out obliquely.

It is a great mistake to load oneself with a host of paraphernalia. The labour of carrying a heavy pack often destroys what might otherwise have been an enjoyable excursion.

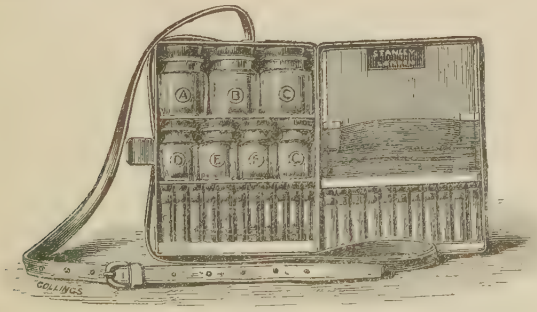


FIG. 118.

A set of half-a-dozen small corked bottles or tubes, and as many small tinned boxes, will complete the collector's outfit. A small but handy pocket collecting case was introduced several years ago by Stanley, of London. (See Fig. 118.)

The objects having been collected, the next thing is to

find out what they are, genus and species, often no easy task for the beginner.

One way of getting over the difficulty is by consulting the books mentioned under each heading—nearly always to be had at the free libraries—often a long, laborious, and unsatisfactory task; while the other is by sending the specimens in a tube to a friend or naturalist of repute. In the event of one not being known, the author has much pleasure in suggesting the name of Mr. Thomas Bolton, who for several years has established a naturalist's studio in Birmingham.* A word to such inquirers—never forget to enclose a stamp for reply: many forget this, and thus the willingness of the naturalist to furnish information gratis becomes a serious tax upon his pocket.

We may now pass on to the enumeration of many objects of interest; but let it not be for a moment supposed that it is possible to give a complete list of objects suitable for microscopic study. The main wish of the author is, to put before the reader something he may collect easily, in the hope that he will become interested in the study of the details of some one of them.

ALGÆ.—The members of this class of Thallophytes may be found almost anywhere, in ditches, streams, ponds, and even in the small pools of water lying in the hoof-prints of animals upon clayey or boggy soils. One of the most interesting of the Algæ is the *Volvox globator* (Fig. 119), which, however, is very uncertain in its habitat. Wherever found it is usually plentiful. All the fresh-water Algæ may be collected by the use of the appliances already mentioned. Many Oscillatoriæ grow upon the surface of the mud at the bottom of pools, and so require the scoop shown in Fig. 114. The whole collection, including mud, should be wrapped up in the muslin, and carried home in that state for examina-

* 57, Newhall Street, Birmingham.

tion. The remainder of the Algæ may be carried home in tubes or bottles, and upon arrival should be emptied into small aquaria formed of wide-mouthed bottles or small propagating glasses turned upwards, the knob resting in a hollow support. Whether minute or not, the gatherings should be examined on the spot with a platyscopic lens, to prevent the loading of one's satchel with useless specimens.

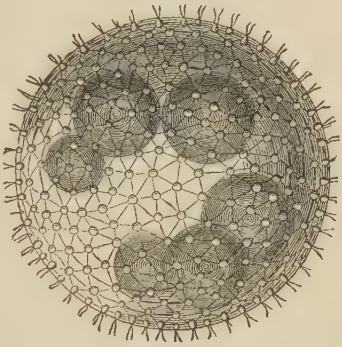


FIG. 119.

Marine Algæ furnish many beautiful objects for the microscope, and can be easily collected upon many shores. Perhaps of all places, Tenby, Torquay, and the Mumbles near Swansea are the best hunting grounds. The various species of Cladophora, Ptilota, Dasya, Bangia, Ceramium, and Griffithsia all form good objects.

Books which may be consulted: Rabenhorst's 'Flora Europæa Algarum'; Griffiths and Henfrey's 'Micrographic Dictionary'; Johnstone and Croall's 'British Sea-Weeds'; Hassall's 'Fresh-Water Algæ.'

ANIMALCULES.—Taking this term to apply to the Infusoria and Rotatoria the student will find a good field for study. It is scarcely possible to find a drop of water which has been for any length of time exposed to the air, not containing either Infusoria or Rotatoria. In some waters they are found in but few numbers, while other localities literally swarm with them. In the former case the pond-filter shown at Fig. 116 will be found valuable. The organisms may be taken from the pond or stream by means of the stick and bottle, and after straining, the residual water carried home in the cork tubes or bottle

which the collector may have with him. A word of advice to the student: Do not overcrowd the organisms, and do not leave any portion of the bottle or tube filled with air if they are to be exposed to shaking or concussion. On arriving home the contents of the tubes may be emptied into small aquaria improvised from broken wine glasses, or—better perhaps—the tube can be stuck through the centre of a large cork and floated in a vessel of water to maintain an equal temperature, when the organisms can be easily abstracted as required by means of a dipping tube.

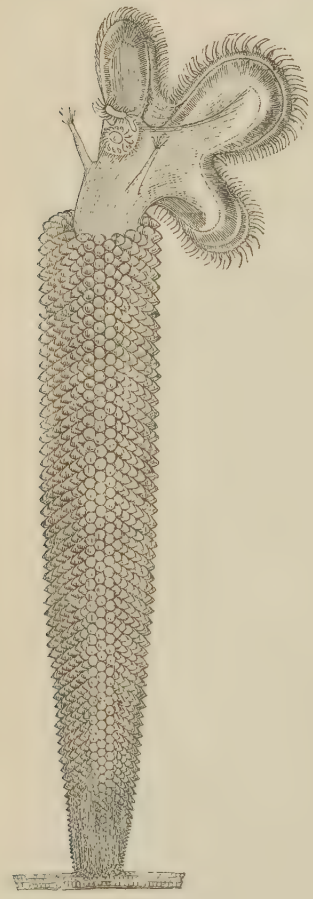


FIG. 120.

Amongst the Infusoria the *Euglena viridis*, *Paramecium aurelia*, and *Coleps hirtus* are good objects for study; of the Rotatoria, *Anuræa longispina* has been found in the tap water furnished by the Birmingham corporation, and others are to be met with in the same habitat, notably *Triarthra longiseta* and *Salpina redunca.*

Melicerta ringens, the tube-building rotifer (Fig. 120) is a beautiful object, and should be carefully searched for; it is frequently found attached to water plants, such as the *Potamogeton crispus*, or large-leaved pond-weed; the *Anacharis alsinastrum* and *Myriophyllum spicatum.*

Books which may be consulted: W. Saville Kent's 'A Manual of the Infusoria'; Griffiths and Henfrey's 'The Micrographic Dictionary'; Pritchard's 'History of the Infusoria.'

ARACHNIDA.—This class of animal life containing the spiders may become very interesting and instructive objects. It seems hardly necessary to say where they may be found, or how to collect them; but it may be necessary to enjoin keeping them moist when they are required for permanent objects or for dissection; diluted glycerine or diluted acetic acid will effect this.

The respiratory system, the circulating system, the spinning organs, and even the eggs, are very interesting, but a knowledge of dissection must be gained before the student can make a successful study of this branch.

The Arachnida are very plentifully distributed—the mites or Acarini, such as the *Acarus domesticus* (cheese mite) and the *Acarus farinæ* (flour mite) are good objects for the $\frac{1}{2}$-inch objectives used binocularly, either alive or when mounted without pressure.

Books which may be consulted: 'Micrographic Dictionary'; Blackwall's 'British Spiders' (Ray Society); Walker's 'British Spiders' (Ray Society).

ANIMAL PREPARATIONS.—The number of these objects is legion; and little else can be done here than to say that animal preparations, as a rule, require special preparation and treatment. Still there is the raw material to collect, and this should be carefully preserved, in order that when examined, its characters shall be faithfully delineated.

The hairs of animals and scales of fish present no unusual difficulties; but such subjects as skin, tongue, liver, lung, &c., should be reserved until the student has become a moderately expert experimentalist. Frogs, mice, rats, rabbits, and guinea-pigs are generally pressed into this service.

Books which may be consulted: 'How to Work with the Microscope,' Beale; Brunton, Foster, Klein, and Sanderson's 'Handbook for the Physiological Laboratory'; Sylvester Marsh's 'Section Cutting'; Rutherford's 'Practical Histology.'

CRUSTACEA.—In this branch are specimens innumerable, the Entomostraca being included under this head. They may all be taken with the appliances already mentioned, and what will do for Infusoria will also be sufficient for Crustacea. *Daphnia pulex*, the water flea; *Cyclops quadricornis; Cypris tristriata; Argulus foliaceus*, the fish louse; *Asellus vulgaris*, the water wood-louse; *Gammarus pulex*, the fresh-water shrimp; *Bosmina longirostris* (Fig. 121);

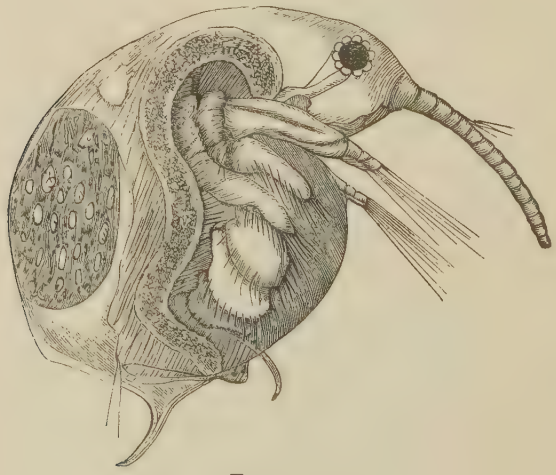


FIG. 121.

Chirocephalus diaphanus, the fairy shrimp, are all to be found in easily accessible ponds during the spring, summer, and autumn.

Books which may be consulted: Baird's 'British Entomostraca'; 'The Micrographic Dictionary.'

DIATOMACEÆ.—This probably has been the most attractive class to nearly all microscopists. Diatoms are a family of Confervoid Algæ, in which the protoplasm is enclosed in silicious valves, generally covered with very fine markings, the nature of which has not yet been satisfactorily made out. They are found in fresh, brackish, and salt water, adhering to plants and stones, or scattered amongst peat, water mosses, or Oscillatoriæ, and even upon damp ground. Nothing is easier than their collection, but of course it is not always possible to meet with the specimens desired.

Diatoms are often found in the stomachs of fish, especially crustaceans and molluscs, and several species have been found in the internal arrangements of *Noctiluca miliaris*, a small exceedingly transparent organism of the size of a grain of mustard seed, causing a phosphorescence in the sea.

A little experience will enable any one to find and to gather all he may desire. Those living in the city can easily procure many beautiful varieties by simply fastening a muslin bag like an umbrella cover to the hydrant. After securing a quantity of the sediment, empty it into a large fruit jar or other receptacle nearly filled with water, and let it settle.

The green, brown, or fawn-coloured scum on the surface of pools, bogs, and marshes, is mostly diatoms, and it may be taken up by means of a spoon or bottle and preserved, always in alcohol and water, or dried upon paper. The living weeds should be taken carefully from their location without much compressing or washing. The finer water plants yield the richest harvest. Fresh-water forms are sometimes found hanging in green-coloured masses from drains, sluices, and water-pipes. To gather from the lake, a net of fine muslin, having an opening in the bottom in which a wide-mouthed phial is tied, may be towed at the

stern of a steamer. The sediment left in the bottom of pails, barrels, and other vessels contains a good supply. To obtain varieties not found at home, open a correspondence with gatherers in other localities, who will gladly exchange.

Dr. Redmayne, in a short communication to 'Science-Gossip' in 1875, described the simple arrangement for diatom collection shown in Fig. 122.

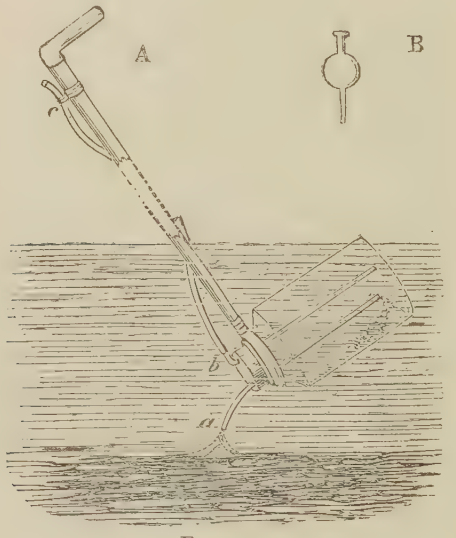


FIG. 122.

A cork must be provided which fits the collecting bottle tightly; this is to be bored with two holes: in each is fitted a glass tube as shown in the figure, one having a slight curve *a*, the other *b* bent at right angles an inch from the end. To the tube *b* is attached a piece of indiarubber tubing about the length of the collecting-stick, and the free end *c* may be held in the hand or fastened to the stick with a small elastic band.

To use the apparatus, the thumb of the right-hand must

press the tube firmly against the stick at *c* and the bottle be lowered until the mouth of the tube is within a quarter of an inch of the diatoms; the thumb is then to be raised, and if the water is deep the bottle will fill by atmospheric pressure, carrying in the diatoms at the same time. In shallow water suction will be necessary to exhaust the air in the bottle, in which case the bulb pipette shown at B in the same figure will be useful as a mouthpiece.

In the collection and recognition of diatoms, the student will find Professor Brown's pocket microscope a useful adjunct, as it is furnished with a deep eye-piece and objectives of an inch, and a fifth of an inch focus. It is shown in Fig. 123.

FIG. 123.

Great care should be taken in the collection of diatoms, so as to have them in as pure a state as possible, as it is not easy to separate them from foreign matter when it is mixed up with them. The late Dr. Redmayne placed the gathering in a long bottle in the sun for a few hours, the lower half of the bottle being covered with black paper. The free diatoms separate themselves from the mud and come to the surface, and can thus be removed.

Fossil Diatomaceæ also exist in immense quantities in various places. The large deposits of guano, the Bermuda deposits, the Berg-mehl in Norway, the Mourne Mountain deposit in Ireland, the recent discovery of diatoms in the London clay by Mr. Shrubsole, and the still more recent discovery in Llyn Arenig Bach, about midway between Bala and Festiniog, in North Wales,

show that the Diatomaceæ are, and have been, very widely distributed.

Some collectors may consider the pocket microscope shown in Fig. 123 not steady enough for general use, and therefore may prefer the form shown in Fig. 124 as made

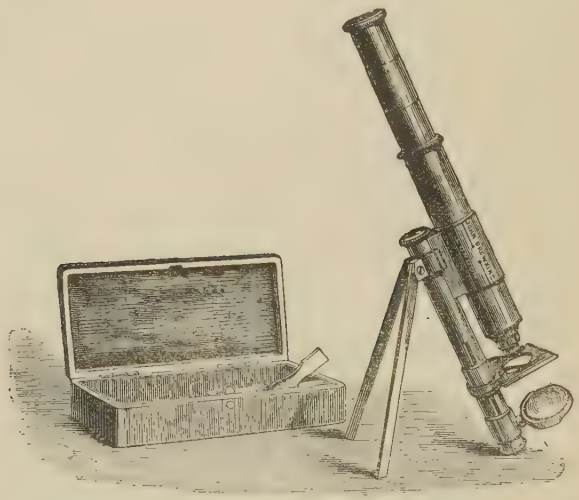


Fig. 124.

by Mr. Browning and others; it is exceedingly portable and very steady, as the author can testify from its practical use when travelling in North Wales.

Amongst the diatoms which may be singled out for examination are *Pleurosigma angulatum* (Fig. 110), *P. formosum* (Fig. 109), *Navicula firma* (Fig. 125), *N. lyra* (Fig. 126), *N. rhomboides* (Fig. 111), *Isthmia enervis*, *Arachnoidiscus Ehrenbergii*, *Meridion circulare*, *Diatoma vulgare*, and a host of others.

Whilst writing this chapter the author has received a tube of diatoms from Mr. Bolton of Birmingham, consisting of a number of species found attached to algæ in the canal of that neighbourhood; the most easily recognised

were:—*Bacillaria paradoxa, Nitzschia sigmoidea, N. lanceolata, Grammatophora marina, Amphiprora alata, Pinnularia radiosa,* and *P. viridis.*

FIG. 125.

FIG. 126.

Books which may be consulted:—Rabenhorst's 'Flora Europæa Algarum'; J. Smith's 'British Diatomaceæ'; Donkin's 'British Diatomaceæ'; and Schmidt's 'Diatomacean Atlas.'

ECHINODERMATA.—The marine objects, star-fishes, sea-eggs, or sea hedgehogs, may always be taken at low water after a spring tide. In their earlier stages they are extremely interesting, being infusoria-like organisms, and often appearing without any internal structure whatever. They furnish the microscopist with abundance of material. The prickles or spines, hooks, and the pedicellaria make interesting and instructive objects.

A section of Echinus spine is used as a test for the flatness of field in low-power objectives, while a section of a small spine may be used for the same purpose with higher

powers. A section of one of these may be seen delineated in Fig. 107.

Books which may be consulted: Forbes's 'British Star-Fishes'; Agassiz's 'Echinodermata Viv. et Foss.'

FERNS require no special apparatus for collecting save a sharp knife and a tin collecting box; they form a very interesting study. The stem may be double stained, as described in the chapter on the staining and injecting of objects.

How many microscopists are there who possess *fern preparations* in their cabinet, spores and sori, stems in cross and vertical section, double and single stained, and yet are totally unacquainted with the life-cycle of a single species; who have looked at sori and spores innumerable, and yet never made the effort of allowing these to germinate, and to observe them in their various and strange mutations.

The *Lastrea filix-mas*, or male fern, is one of the best species to study for the beginner, and there are many others easily found, such as *Athyrium filix-fœmina* (lady fern), *Scolopendrium vulgare* (hart's tongue), *Pteris aquilina* (common bracken), *Adiantum capillus-Veneris* (maiden hair), and many others.

Books which may be consulted: Moore's 'Index Filicum'; 'Handbook of British Ferns'; Newman's 'British Ferns'; J. Smith's 'Ferns, British and Foreign'; Hooker and Baker's 'Synopsis Filicum.'

FORAMINIFERA.—These gelatinous, structureless animals are mostly sought after for the sake of the shells, serving them as a covering. The shells are pierced with holes, through which the animal protrudes its pseudopodia, using them as a means of locomotion. They are found in largest numbers in the sand and mud from the sea-bottom, but may also be found on sea-weeds, and in

the fossil state in chalk, limestone, and other mineral deposits. From sea-soundings they may be procured by dissolving out the tallow in which they are collected, by means of benzine—any sort of benzine or benzoline serving this purpose. To obtain fossil Foraminifera from chalk, the pieces must be broken up small by means of a hammer, and then gently crushed in an iron mortar. The powder is then to be placed in a piece of coarse calico, tied up like a pudding, and put into a large basin of water, and well kneaded, until reduced to one-third its original bulk. The milky fluid is then to be poured off, until only one-fourth remains, and the operation of washing, by stirring up with

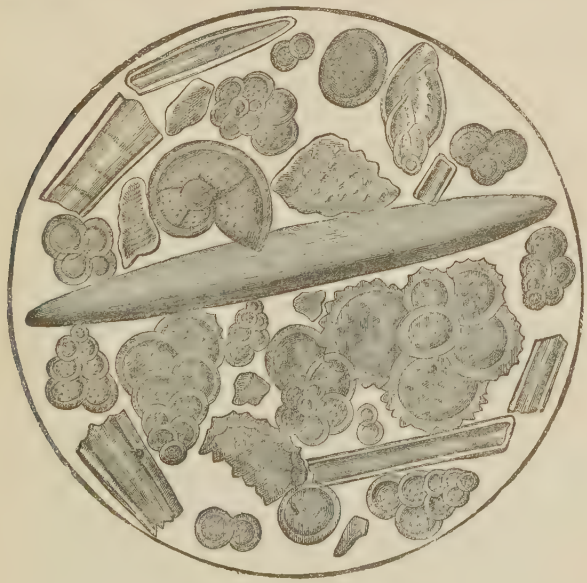


FIG. 127.

fresh water, and allowing to settle, repeated many times, until a portion of the residue, on examination under the microscope, shows that most, or all, of the extraneous matter has been eliminated.

The illustration Fig. 127 shows a section of chalk from Gravesend with Foraminifera *in situ.*

Amongst others, *Lagena squamosa, Orbitolites complanatus, Polystomella crispa,* and *Nodosaria raphanus,* are very good objects.

Books which may be consulted: Williamson's 'Recent Foraminifera' (Ray Society); Carpenter's 'Introduction to the Study of Foraminifera.'

FUNGI.—Micro-fungi may be found everywhere, and make a splendid study. Many of them, however, can only be examined successfully when in the fresh state, such as *Penicillium crustaceum* and *Aspergillus glaucus,* the common moulds and mildews of our houses. Nearly every plant and tree is attacked at one time or another by some particular species of micro-fungus, so that the student will find plenty of work in this class alone. We have the *Puccinia graminis* upon the leaves and stems of standing corn, as well as *Tilletia caries* and *Ustilago segetum* or smut, which fill up and destroy the whole contents of the ear; the *Æcidium* on the berberry bush; *Triphragmium* on the leaves of the meadow-sweet; the blackberry brand, *Aregma bulbosum; Coleosporium synantherarum* on the colt's-foot; *Cystopus candidus* on cabbages; *Peronospora infestans* on our potatoes; *Peronospora gangliformis* on lettuces; and *Peronospora viciæ* on peas: a host of others being within easy reach.

The micro-fungi shown in Fig. 128, are as follow:—

a. Stachybotrys lobulata.
b. S. atra.
c. Penicillium sitophilum (*Oidium aurantiacum*).
d. Myxotrichum deflexum.
e. Polyactis fasciculatis.
f. Sporocybe alternata (*Aspergillus alternatus*).
g. Rhopalomyces pallidus.
h. Papulaspora sepedonioides.
i. Acremonium alternatum.

They have all been found upon moist cotton goods or in analogous situations.

All that is necessary in collecting micro-fungi is to well isolate each specimen, preferably by wrapping in soft paper, and placing separately in a small box.

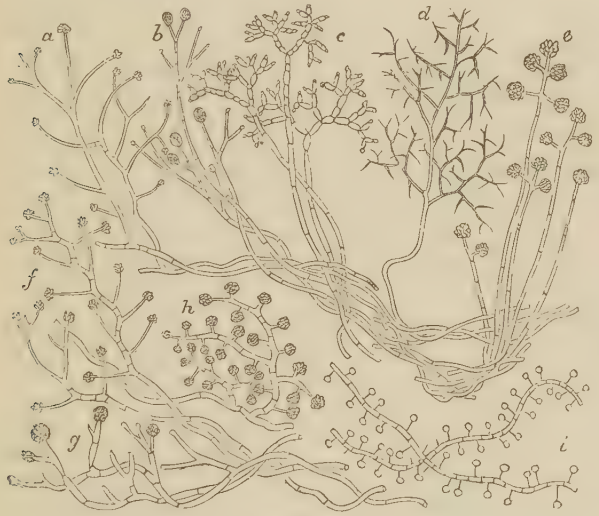


FIG. 128.

Books which may be consulted: 'Outlines of British Fungology,' by Rev. M. J. Berkeley, F.R.S.; 'Handbook of British Fungi,' by M. C. Cooke; 'Rust, Smut, Mildew, and Mould,' by M. C. Cooke; 'Selectæ Fungorum Carpologia,' 3 vols., Tulasne.

INFUSORIA AND ROTATORIA.—Nearly all that is necessary to say under this head has been given when treating of animalcules. Fresh-water infusoria may be collected in wide-mouthed bottles, and the individuals selected for examination removed by means of one of the dipping tubes shown in Fig. 117. The members of this class of animals are generally to be found attached to weeds. The beautiful

Stephanocerus Eichhornii is one of this class; it is shown in Fig. 129. The author has before him at the present moment some leaflets of *Myriophyllum spicatum*, the common water-weed, completely covered with *Melicerta ringens*, *Floscularia cornuta*, *Philodina megalotrocha*, *Limnias ceratophylli*, *Euchlanis dilata*, and *Codosiga botrytis*. Most of these organisms may be obtained from Mr. Bolton's studio.

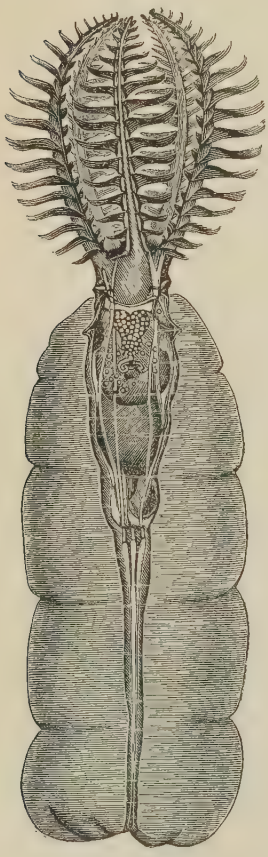


FIG. 129.

Books which may be consulted (see Animalcules).

INSECTS afford an inexhaustible series of treasures to the microscopist; but in order to examine these perfectly, a knowledge of dissection must be gained. The head, eyes, legs, wings, proboscis, spiracles, and tracheæ, all make instructive objects. Many are the kinds of insects that can be pressed into the microscopist's service, the *Siphonoptera*, or fleas; the *Parasitica*, or lice; the *Diptera*, or flies; the *Hymenoptera*, bees and wasps; *Lepidoptera*, butterflies and moths; *Orthoptera*, grasshoppers and crickets; the *Coleoptera*, or beetles, are all too well known to require instructions when, how, and where to collect.

Fig. 130 shows the male *Dytiscus marginalis*, or great water beetle, a very interesting insect to the microscopist, while Fig. 131 is a picture of the female.

The larvæ too are an interesting study; this stage in the gnat, dayfly, and even of the *Dytiscus*, Fig. 132, will amply repay the observer for the attention he bestows upon it.

FIG. 130.

FIG. 131.

When required for immediate examination, no special care is requisite; but if for dissection and permanent mounting afterwards, they should be immersed in a mixture of equal parts of glycerine and water, dilute acetic acid, or even dilute alcohol (for some things which require hardening), in order to ensure preservation. It must be understood, however, that this is not universally applicable. There are some objects which would be spoiled by immersion in a fluid, and these, of course, must be prepared while in the fresh state.

FIG. 132.

The scales of several insects are much used as test objects, notably those of the *Lepisma saccharina*, *Morpho menelaus*,

the *Polyommatus argus*, the *Lepidocyrtus curvicollis*, and *Podura plumbea*. These scales are shown in Chapter V. The *Podura plumbea* is a small insect found in damp shady places, under stones in cellars, where they may be caught by sprinkling a little oatmeal near their haunts. They are commonly called spring-tails; and in order to help the student in finding them, an illustration is given in Fig. 133, and of the *Lepisma saccharina* in Fig. 134, each magnified about 12 diameters.

FIG. 133.

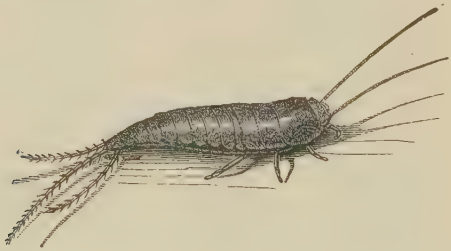


FIG. 134.

It is very interesting at times to watch the development of insects and the changes they undergo during their cycle of existence; for this purpose a "vivarium" is necessary, so that while furnishing them with sufficient air they may be prevented from crawling or flying away, and the respired carbonic acid find easy exit.

The best kind of vivarium is that shown in Fig. 135, and is so simple that an ordinary tinman can construct one for a trifling cost.

The framework is made of perforated zinc with a bottom of the same material, on which rests an inner circle of zinc but perforated more closely. This is not fastened to the bottom

but can be raised at will; it is covered with a round piece of flat glass. On this stands a saucer of well moistened clay stuck round the edge with such leaves as the insects feed on. In the centre of this saucer is a small vase filled daily with fresh flowers and grasses for the butterflies.

Fig. 135.

It is easy in this apparatus to raise moths, butterflies, and other winged insects, such as saw-flies and the like, from the egg; and the many changes may be watched through the glass, from the emergence of the caterpillar from the egg to the exit of the imago from the pupa case.

Lichens.—These interesting vegetable productions require but an old knife and a few pill-boxes for their collection, with occasionally a hammer and chisel. The purer the atmosphere of the district the greater variety to be found, while in a smoky and gas-polluted atmosphere they are either not found at all, or only to be met with in the gonidial state, when they may be easily mistaken for protophytal algæ. Lichens are to be found growing

upon rocks, stones, trees, old railings, twigs, bushes, heath, moss, and in many other places.

Lichens are mostly of large size, and require thin sections to be cut ere the structures can be satisfactorily made out; but some should be always mounted as opaque objects, to show what the natural form of the plant is.

FIG. 136.

FIG. 137.

Fig. 136 shows a piece of fencing upon which is growing *Lecanora cerina*, while the next illustration, Fig. 137, shows *Usnea barbata*, one of the filamentous species. Among the more familiar kinds are *Parmelia parietina*, *Lecidea*

canescens, Physcia ciliaris, Cladonia pyxidata, and *Pertusaria communis.*

Books which may be consulted: 'History of British Lichens,' W. L. Lindsay, M.D.; 'Lichenes Britannici,' Crombie; 'The Lichen Flora of Great Britain and Ireland,' Rev. W. A. Leighton.

MINERALS.—Of late years much has been discovered in the mineral kingdom by the aid of the microscope, but owing to the nature of the substances to be examined, much preparatory work has to be done ere a satisfactory examination can be made. Sections have to be cut and semi-polished ere the structure can be well made out, and this will be described in our next chapter. Barbadoes rock showing Polycistina *in situ,* oolitic and nummulitic limestone, slags from iron and copper furnaces, fossil wood,

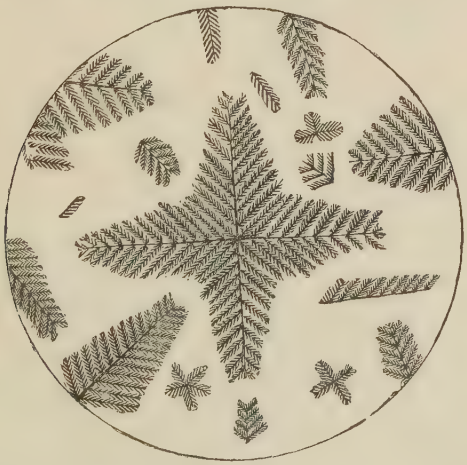


FIG. 138.

minerals used in smelting, basalt, boiler incrustations, coal, shale, lava, and many other minerals, furnish instructive objects, and may be collected in plenty in the proper localities.

A section of porphyrine is shown in Fig. 138, which

forms an illustration of the production of crystalline forms when molten vitreous matter is allowed to cool slowly. This mineral is of a beautiful crimson colour, and under a binocular microscope few objects can vie with it for beauty.

FIG. 139.

The two figures, 139 and 140, show the artificial production of similar crystals during the manufacture of glass. When the "metal" is cooled slowly, crystals of certain forms are obtained, which vary in form according to the differences existing in the quantities of the materials from which the glass is made.

Books which may be consulted: 'Sur les Crystallites,' Vogelsang; Allport on the "Microscopical Examination

FIG. 140.

of Rocks and Minerals," p. 98, 'Monthly Microscopical Journal'; 'Mineralogy and Study of the Rocks,' Rutley.

MOLLUSCS.—The entire series of the Mollusca form very

interesting objects for the microscope, the structure of the viscera, the spermatozoa, and the tongue or *odontophore* being the chief. Fresh-water and land snails should be avoided by the student until he has gained sufficient experience in the manipulation of limpets and whelks, from which the extraction of the *odontophore* is a comparatively easy operation. The marine molluscs may be carried long distances folded in damp sea-weed, while fresh-water snails should be conveyed in a similar manner in damp Anacharis. When the animals are not immediately required, they may be dropped into glycerine, dilute acetic acid, or into alcohol. Edwards advises that they be dropped into caustic potash solution (say one part of caustic potash and two of water), until they begin to decompose; but of course this is only admissible when nothing but the *odontophore* is required.

This odontophore is the so-called tongue or lingual ribbon, really the masticatory apparatus, a long ribbon-like organ furnished with a complicated system of teeth, generally set upon flattened plates. The form and arrangement of the teeth furnish characters of much importance in classification, and therefore should be studied by all those who are interested in this department of natural history.

FIG. 141.

Fig. 141 shows a short length of the odontophore of the common limpet, *Patella vulgata.*

Books which may be consulted.—'British Mollusca,' Forbes and Hanley; "Notes on Victorian Molluscs and their Palates," C. M. Mapleston, 'Monthly Microscopical Journal,' vol. viii. p. 45; Alcock, 'Proceedings of Manchester Literary and Philosophical Society.'

MOSSES.—These require but few instructions for collecting. They are of little use for the purposes of scientific inquiry without fruit, and therefore the collector should always endeavour to procure the plant in fructification. *Polytrichum commune, Sphagnum squarrosum, Hypnum purum, Tortula* (*Barbula*) *muralis,* are amongst the more common species.

Every month produces some species to be collected; on old walls, the roofs of tiled and thatched buildings, on our garden walks, on our downs and commons, on chalk and sandstone, in bogs and on the mountain-top, mosses may be found everywhere. The necessary apparatus for collecting consists of an old pocket-knife, a platyscopic lens, as shown in Fig. 2, a satchel or small bag, and a good supply of thin envelopes, or square pieces of tissue or other thin paper, in which to wrap each species.

Books which may be consulted.—Hooker, Taylor, and Wilson's 'Bryologia Britannica'; R. Braithwaite's 'British Moss Flora'; Stark's 'Popular History of British Mosses'; Berkeley's 'Handbook of all British Mosses'; Unwin's 'Dissections and Illustrations of British Mosses.'

POLYCISTINA.—These animals are a family of Rhizopoda Radiolaria, the silicious skeletons of which form a series of very interesting objects for the microscope. They exist on every ocean floor and embedded in rocks, fossil Polycistina having been found at Oran, Bermuda, and Barbadoes, as well as in many other places. The silicious skeletons, or shells, as they are sometimes termed, with their prolongations, are aids in discriminating between Polycistina and Foraminifera, which former some of the species resemble. A section of Barbadoes rock, showing Polycistina *in situ,* is often a pretty, interesting, and instructive object. These objects may be obtained from Barbadoes earth, by boiling with its own weight of washing

soda and a similar weight of water for one hour. After allowing to settle, the "flock" must be poured off and the boiling with soda repeated, with intermediate washing several times. *Rhopalocanium ornatum*, *Podocyrtis Schomburghii*, and *Astromma Aristotelis* are, perhaps, the most beautiful of this class.

Books which may be consulted: Haeckel's 'Die Radiolarien'; 'The Micrographic Dictionary.'

POLYZOA.—These interesting objects have, by the researches of modern inquirers, become much more known; they are both marine and fresh-water, and may be found in many rivers and canals.

On the sea-shore they are very plentiful, forming a crust on submerged rocks, or attached to stones, shells, and sea-weed. The individual animal is termed a *polypide*, and the colony a *cœnœcium*, which latter consists of an aggregation of cells or cups, often taking elegant and vegetable forms. Most Polyzoa are fixed organisms, but one at least—the *Cristatella mucedo*—is capable of locomotion.

One beautiful form, the *Lophopus crystallinus*, may be found constantly in the Gorton Canal, near Manchester, and is much sought after by microscopists, visitors to the neighbourhood; it is shown in Fig. 142.

The study of these animals is an interesting one. In *L. crystallinus*, as shown in the figure, *a* is the region of the mouth; *b*, the œsophagus; *c*, the stomach; *d*, the intestine; *e* are the retractory muscles; *f*, a statoblast; *h*, the mouth; *i*, tentacles retracted, partly within cell; *k*, the outer transparent envelope; *l*, the perigastric space; *m*, the lophophore; and at *n* the tentacles have been excised, to show mouth.

Of marine Polyzoa, the sea-mat, or *Flustra foliacea*, may be taken as a type; it is most plant-like and flexible. The cells are narrow at the base and rounded at the end, with

scattered marginal spines; when rubbed betwixt the fingers it exhales an odour resembling oil of lemon. An illustration of this common object of the sea-shore is given at

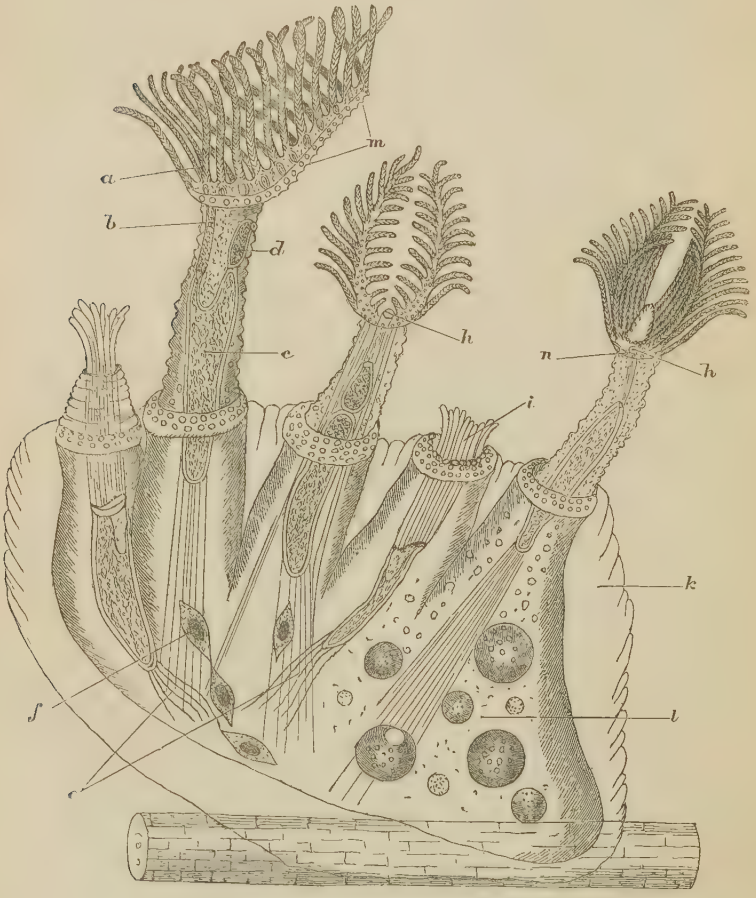


FIG. 142.

Fig. 143, while in the next figure (144) is shown a portion, much enlarged.

When collected, they must be preserved in bottles or

tubes with some of the water they were taken from, while it has been stated that they may be preserved with expanded tentacles by dropping alcohol into the water; but this is not always successful.

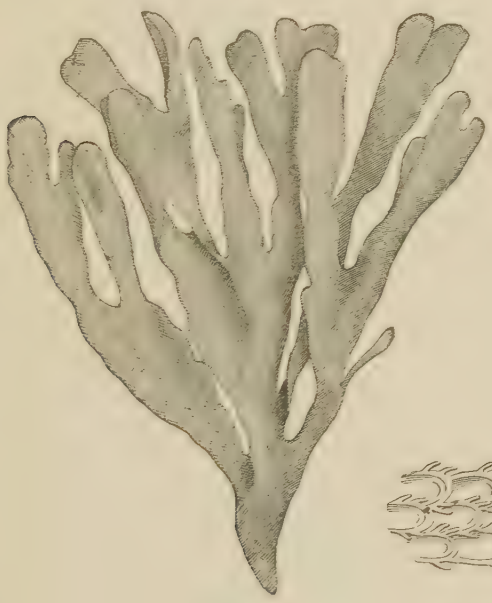


FIG. 143.

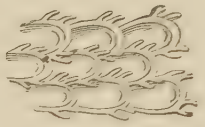


FIG. 144.

Books which may be consulted: W. Saville Kent's 'Manual of the Infusoria'; Johnstone's 'British Zoophytes'; 'Micrographic Dictionary'; Allman, 'Fresh-water Polyzoa' (Ray Society).

VEGETABLE PREPARATIONS.—The raw material for this branch of study may be found everywhere, the vegetable kingdom being able, perhaps, to supply more objects for the microscope than all the other classes together. Algæ, diatoms, ferns, fungi, lichens, and mosses, have already been mentioned, but the phanerogams alone are able to furnish matter for an immense amount of observation. The

stems of trees and shrubs, the pine, fir, hornbeam, box, birch, beech, oak, chestnut, and elder, being easily accessible, are interesting when cut into sections. The leaves, seed, pollen, roots, hairs, cuticles, raphides, and a host of other preparations, may be obtained from almost any plant, whether it be a choice specimen from the greenhouse or a common weed from the garden.

The starches form a study in themselves, and are specially interesting from many points of view; they are easily prepared and should be kept dry, in small homœopathic tubes, for observation. The grains, such as wheat and barley, may be bruised and set aside for a few days to ferment, when, upon squeezing through a fine cotton cloth, the milky liquid contains the starch. Roots, rhizomes, and tubers require to be rasped, and the milky fluid strained in the same manner. After a time, the starch granules subside, when the supernatant fluid must be poured off and fresh water added, the whole stirred up, and allowed to settle again. After pouring off the water, the granules should be allowed to drain upon blotting-paper and dried at the temperature of the air in a warm room. Fig. 145 shows several of the commoner forms of starch.

Nothing special is required for the collection of raw material for vegetable preparations, without it be a tin case, for holding leaves, stems, or the whole plant, and squares of soft tissue paper, in which to carefully fold specimens. In collecting pollen, it is well to gather the complete flower, and carefully isolate the specimens in order that no confusion of the granules may take place. The pollen of a species of the mallow tribe is often used as a test object for low-power objectives. It is shown in Fig. 97.

Works which may be consulted: Maôt and Decaisne's

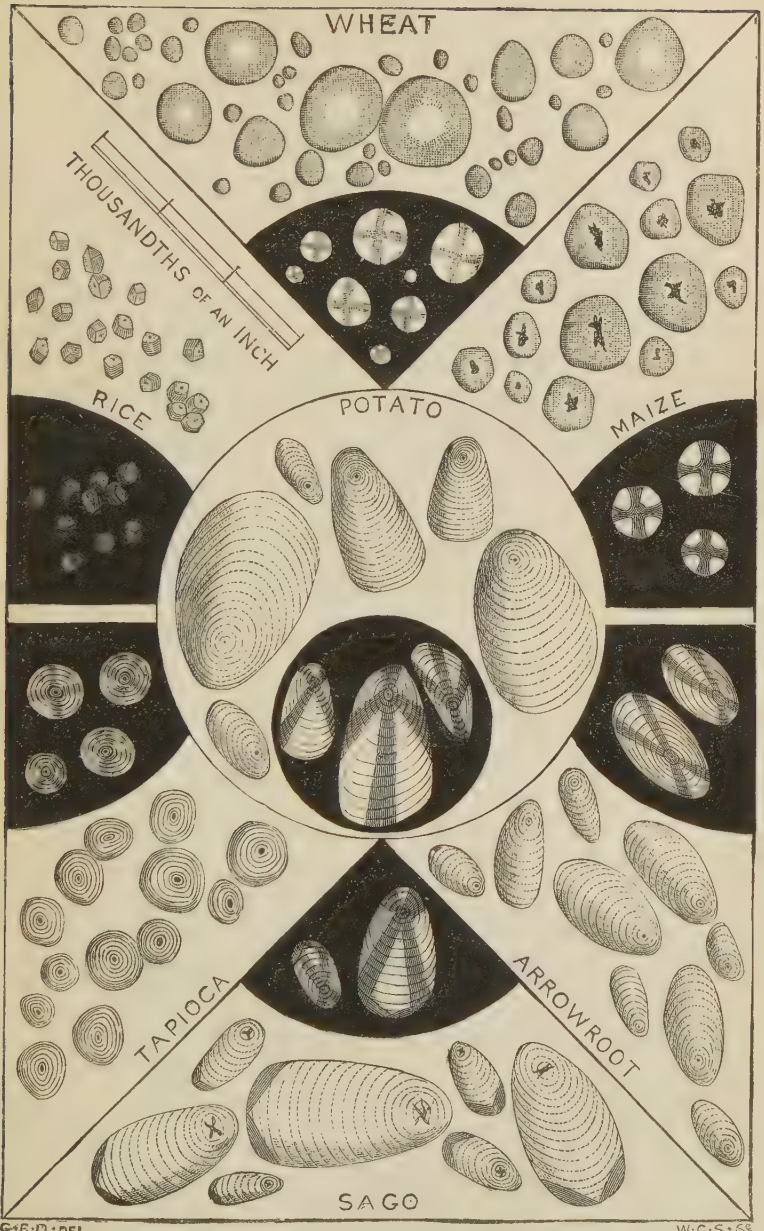

WHEAT
THOUSANDTHS OF AN INCH
RICE
POTATO
MAIZE
TAPIOCA
ARROWROOT
SAGO
G·E:D·DEL.
W·G·S·Sc

FIG. 145.

'Descriptive and Analytical Botany,' by J. D. Hooker; Sachs' 'Text-Book of Botany' (Clarendon Press); Balfour's 'Manual of Botany.'

ZOOPHYTES.—This division includes the Hydrozoa and the Actinozoa, both of which furnish many objects for study. In the first class we find the *Hydra viridis* and *H. fusca*, which may be collected without much trouble from streams and pools; it is visible to the unassisted eye, though it can only be satisfactorily examined by the aid of the microscope. Fig. 146 shows *H. viridis* as it usually occurs attached to duckweed, and in the same manner it is often found on *Anacharis alsinastrum* and other water plants.

FIG. 146.

Another order of the Hydroid Zoophytes is the Tubularida, of which *Tubularia ramea* is a specimen. It is commonly called the "branched pipe coralline," and is a marine organism; in fact, all the Tubularida are marine with the single exception of the genus Cordylophora, which inhabits fresh water. The *Tubularia ramea* may be seen in Fig. 147.

Amongst the Actinozoa are the sea-anemones, the coral polypes, and the Cydippe (*Pleurobrachia pileus*).

The life-history of the zoophytes is extremely interesting, and the student will scarcely be able to appreciate that the ovoid ciliated mass, seen escaping from the ova of the parent, would ever lose its cilia and develop into the parental form.

FIG. 147.

When it is desired to keep them for preservation and mounting, the same method may be employed as for Polyzoa, to drop alcohol into the water in which they have been collected, which is stated to kill them while causing but little retraction of the tentacles.

Books which may be consulted: 'The Micrographic Dictionary'; Johnstone's 'British Zoophytes'; Hincks' 'British Hydroid Zoophytes.'

CHAPTER VII.

Micro-Dissections.

Many of the objects mentioned in the preceding chapters cannot be examined under the microscope without subdivision or previous preparation, either on account of their size, or because the parts lie in different planes, the transparent portions being hidden from view by others of an opaque character.

This is so with most insect life, especially the larger species, though, on the other hand, the more minute kinds, such as the *Acarus domesticus*, or cheese-mite, are generally so transparent as to require no special treatment.

Beetles and other insects, flies, snails, frogs, and newts are very good examples of the former class, and the student will do well to practise the dissection of these objects. He will find it absolutely necessary to acquire a good knowledge of dissection, in order to gain a correct insight into the relative structure of the insect economy.

We can hardly describe in words how to proceed in each individual case. Experience will come by practice, and the student will find that each subject becomes more and more easy, especially if, before commencing in haste with the needles and scissors, he will study the general arrangement of organs in his subject, by reference to some one or other of the many standard works in existence, obtainable at any free library.

One remark, however, is necessary. Do not hurry on with imperfect dissections of a host of subjects; stick to

one insect until a good knowledge of its anatomy has been gained, and by no means take the trouble to prepare and mount any imperfect specimens. Persevere until the dissections are perfect, and then spare no trouble or pains in preserving them for future reference.

Dissections may be carried on with but few instruments; a few small-sized troughs, knives, scissors, needles, and camel-hair pencils are nearly all the requisites. The dissecting microscope must be left to the taste of the operator, who will find many forms to select from.

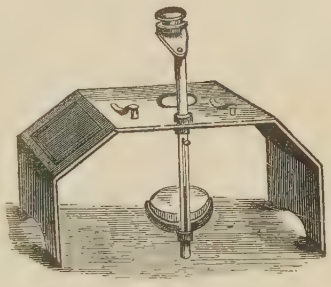


FIG. 148.

Fig. 148 shows Collins' cheap dissecting microscope, of which a large number are in use in Professor Henslow's botanical classes; while the next figure (149) is an illustra-

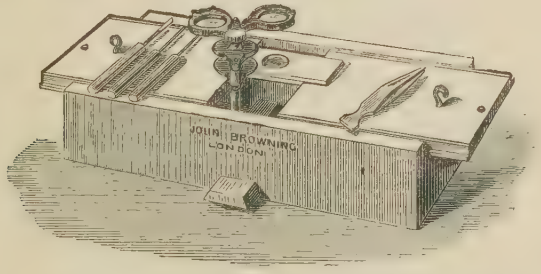


FIG. 149.

tion of Houston's botanical dissecting microscope, which may suit botanists who wish for nothing more than a steady stand for a pocket magnifier.

This ingenious little instrument is intended to provide working botanical students with a convenient and serviceable dissecting microscope at a moderate cost. The box measures, when closed, 9 inches long, 4 inches wide, and

2 inches deep, and is so constructed that, by using a divided sliding lid (acting as a support for the dissecting stage), a rest for the wrists is secured while the hands are employed in dissecting.

The duplex lens, giving three powers, 4, 6, and 10 diameters, is screwed to the end of a brass focussing tube moving upon a brass pillar attached to a sliding bar at the bottom of the box. The lens may at any time be unscrewed and carried in the pocket.

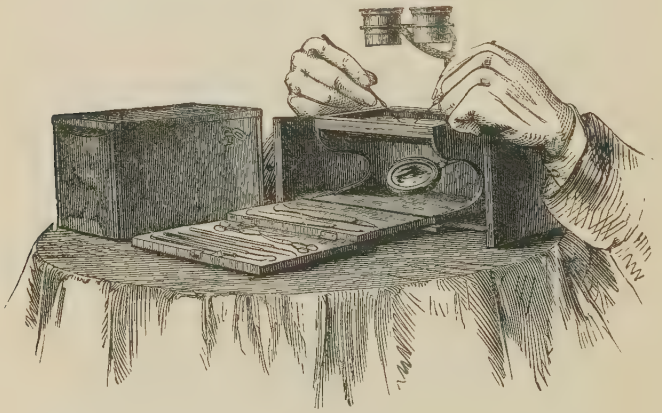


FIG. 150.

The dissecting stage is a cork slide, plain on one side for general work, but provided with a shallow cell on the other, for the dissection of such objects as small glossy seeds, which "fly" under the needles. A pitted glass slide, to be used when the object is best dissected under water, is also provided. A cutting needle, two dissecting needles, and a pair of small forceps are also included, and the whole is sold at the moderate price of 6*s.* 6*d.*

Fig. 150 is a representation of Lawson's dissecting microscope as made by Mr. Collins—a very handy and compact instrument, containing, as it does, in the unfolded case the knives, needles, and scissors for the operator's immediate use.

Recognising the advantages of binocular vision for the purposes of dissection, as well as the erection of the image, Mr. Baker and also Messrs. Swift and Son have introduced a cheap form of the Stephenson binocular, as shown in Fig. 151, the details of which have already been described on page 35.

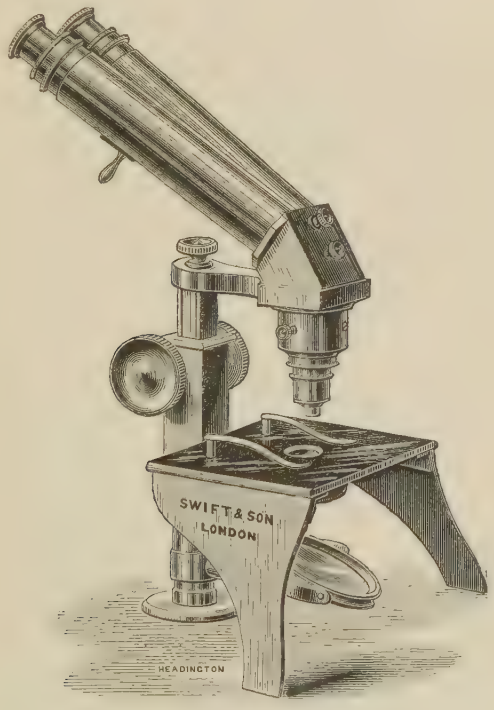


FIG. 151.

If the erection of the image be desired, it can be obtained by the use of the draw-tube and erector already illustrated by Figs. 32 and 33, and this method has many advantages over the use of the Stephenson binocular. The erector screws into the end of the draw-tube, and using the A eye-piece with a $\frac{1}{2}$-inch objective any amplification from 2·5 to 60 diameters may be obtained, or from 12 to 260 diameters

with the D ocular. It must not be forgotten, however, that the use of the erector disturbs the corrections of the objective, producing spherical and chromatic aberrations. This is so great that, when using the A eye-piece and a ½-inch objective, an object may be brought to an indifferent focus at any distance between 1 inch and 2½ inches from the front lens upon the instrument shown in Fig. 24; but at 1½ inches perhaps the sharpest image is formed. The way to remedy the effect of these aberrations is to use the C or D eye-pieces, when the object runs in and out of focus very sharply, and as the peripheral rays are not gathered in, there is but little aberration of either kind visible.

The following table will show the use of the erector and draw-tube, used upon the stand in Fig. 24 with a ½-inch objective of 50° air angle.

Position of Draw-tube.	A Eye-piece.			D Eye-piece.		
	Distance of Front Lens from Object.	Size of Object in Field.	Amplification in Diameters.	Distance of Front Lens from Object.	Size of Object in Field.	Amplification in Diameters.
	in.	in.		in.	in.	
Full home.. ..	1·30	0·75	2·5	1·05	0·40	12·0
Withdrawn ½ inch	0·60	0·40	7·0	0·44	0·15	30·0
,, ½ ,,	0·36	0·20	11·0	0·36	0·09	50·0
,, 1 ,,	0·23	0·12	20·0	0·23	0·05	90·0
,, 2 ,,	0·17	0·06	40·0	0·17	0·03	180·0
,, 3 ,,	0·15	0·04	60·0	0·15	0·02	260·0

For cheapness, there is nothing better than the watchmaker's or engraver's eye-glasses. These may be fixed into any form of stand the ingenuity of the student may devise, and with these alone very much work can be done, quite as much perhaps as he will be able to execute for some time.

The subjects for dissection are usually operated upon under water or some other fluid, in dilute alcohol, or even in glycerine. This is a very convenient medium in which to

carry on minute dissections, as the operation can be performed with much more certainty than is the case in more mobile fluids.

In whatever fluid the specimen for dissection be immersed, a trough is necessary to hold it, such as shown in Fig. 152, and this may be either of guttapercha or glass, not too large, but roomy enough for working, and also for the disposal of loose matter. Half an inch in depth is ample (except for special subjects), as if deeper, the sides are apt to interfere with the free use of the needles and fingers.

If the trough is of glass, a suitable bedding may be run in to a depth of a quarter of an inch, to which the subject can be pinned down. A translucent bottom may be made of a mixture of napthalin and stearine, an opaque bedding of equal parts of resin and beeswax, thinned to the necessary consistency with unsalted lard, and coloured black with lamp-black, or white with oxide of zinc or dried china-clay.

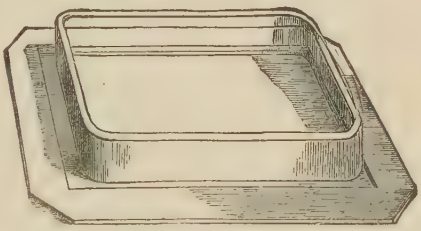


FIG. 152.

While dissecting, no instruments are more handy than ordinary sewing needles, thrust with the eye first into cedar-wood handles. They may be bent to any required shape while heated in the gas or a spirit-lamp flame, and afterwards rehardened by heating and plunging, while hot, into oil. They must be kept well polished, as if any roughness prevail on their surface it is next to impossible to produce anything but torn and disfigured dissections.

Camel-hair pencils are indispensable agents for the

removal of loose matter, torn and unrequired portions of the insect anatomy, and also for the arrangement of the dissected portions.

The knives used in dissecting should be of good steel, and kept in order by the occasional application of a Washita oilstone. Some of the most useful forms are shown in Fig. 153—two or three will perhaps be selected; the author prefers the forms shown at *a*, *b*, *d*, *f* and *h*.

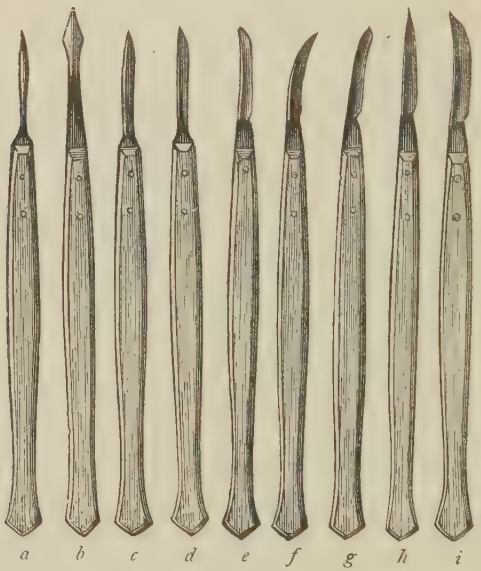


FIG. 153.

Nearly every microscopist prefers to work his own way, and with his own choice of instruments. The spear-head needles sold by Mr. Ward of Manchester will be found exceedingly useful.

Three kinds of scissors are generally required by the dissector, the curved, Fig. 154, elbow, Fig. 155, and plain, Fig. 156. They must be well made, and kept carefully cleaned and free from rust. The application of mercury ointment

will usually go far in protecting steel instruments from this enemy.

Another pair of scissors is also much used in minute dissections, one leg of which is fixed in a light ebonite handle,

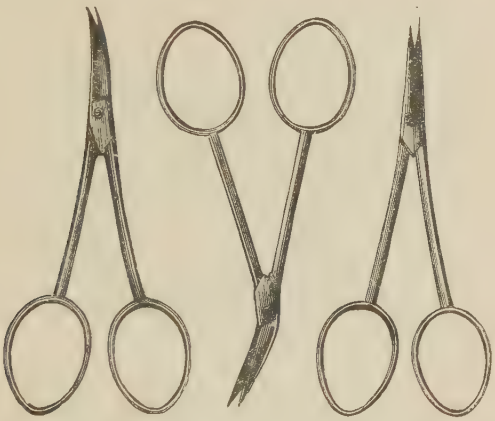


FIG. 154. FIG. 155. FIG. 156.

the blades being kept apart by means of a spring. The pressure of the finger on the loose leg causes the blades to close, and it will be found after a little practice that the instrument may have many applications. It is shown in Fig. 157.

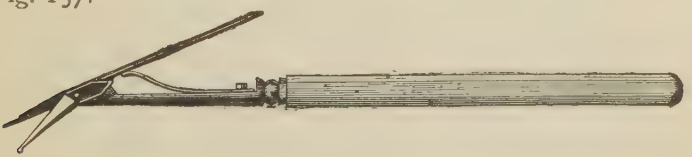


FIG. 157.

The steel forceps, shown in Fig. 158, are necessary adjuncts to the dissector, and he should be careful to select those of but medium strength in the spring, and such as can be depended upon for seizing an object finely yet firmly, and not at all liable to twist aside and spoil the subject under dissection. A pair of curved

forceps, as well as a pair of the straight variety, with fine points, are necessary.

A bull's-eye condenser for concentrating light upon the object, and a small glass syringe for washing subjects and removing or adding to the liquid contents of the trough, complete the dissector's outfit.

And now having his apparatus ready, the student will naturally be looking about for something to dissect. He need not seek long at any season of the year, but in the

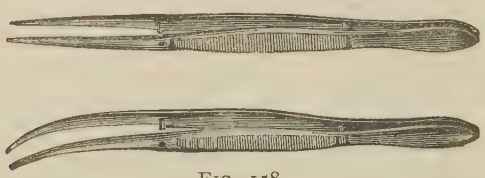


FIG. 158.

event of his not being able to fix on what he shall commence with, the blow-fly, *Musca vomitoria,* is suggested.

Plenty of work will be found in the blow-fly if it be done properly, and after this the student may take in hand the water-beetle, *Dytiscus marginalis;* the grasshopper, *Locusta viridissima;* or the common house cricket, *Acheta domestica.*

In the above, the student will find the tongues, antennæ, eyes, wings, legs, segments, spiracles, and tracheæ, an interesting study, and besides these organs, gizzards will be found in the last three.

The insect (if not too large) should first be held in the stage forceps as shown in Fig. 66, or in any other convenient manner, and examined with a low power (say a 2-inch objective), to show its general character, and give an insight into the manner in which the appendages should be dealt with.

The proboscis of the fly may be obtained by pressing the thorax, so as to cause protrusion, placing the organ

upon a strip of glass, covering with a thin square cover, and severing with a sharp knife. If the insect has been killed with chloroform, the organ will generally be found protruding. Just a word *en passant* to encourage the student to display his dissections naturally, in order to give observers an idea of the real use of the various organs in the insect economy. This cannot be better illustrated than by reference to this proboscis, one of the late Mr. Topping's favourite preparations. The more natural condition should be mounted in glycerine, and though not so pretty an object, is at least truthful.

Put no insect to pain. Kill it at as early a stage in the inquiry as is possible, either with chloroform, bruised laurel leaves, or by means of the cyanide of potassium bottle now so often used.

Proceeding with the blow-fly, the wings may be detached from the thorax by means of the knife, scissors, or forceps, the legs taken off at the thigh, the halteres or poisers detached, and the antennæ cut from the head, completing the list of appendages. The carcase must now be pinned down in the trough, or fixed to the bedding by warming a spot with a hot iron, fixing the subject into the melted stratum, and the integument carefully slit up with a fine pair of scissors, upon both sides. The chitinous skeleton must then be raised with the forceps, and the attachments cleared away with the aid of a blunt needle and a spear-headed instrument, when, if tolerably well performed, the whole of the organs will be seen *in situ.* The subject should now be left in a mixture of glycerine and water (equal parts of each), for about twelve hours, after which treatment the organs may be readily dissected. Dilute alcohol is very useful when dissecting the nervous system of insects. A more or less prolonged immersion hardens the nerve fibre, which the student will find requires very

delicate handling. It is not difficult to find and take away the tracheal system (though the beginner will find this easier to prepare from the silk-worm or caterpillar), and the muscles which are used for raising and depressing the wings during flight may be found without much trouble.

FIG. 159.

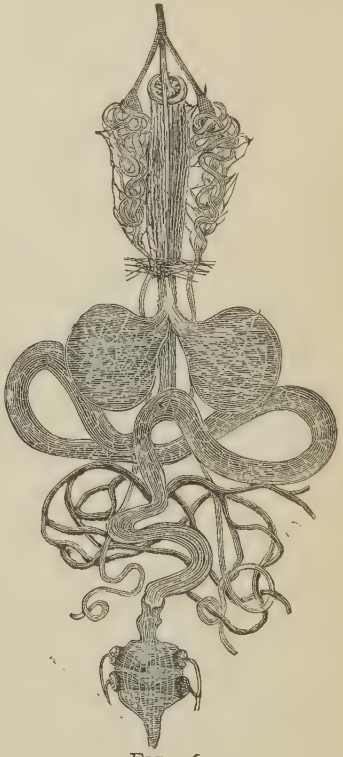


FIG. 160.

The digestive apparatus varies somewhat in different insects, and as an aid to the student the arrangement of these organs is given as they appear *in situ* in the water-beetle (Fig. 159) and the blow-fly (Fig. 160).

The fly lives by suction, and therefore the food does not require any unusual amount of mastication. It therefore

has no gizzard. The water-beetle, on the other hand, has a gizzard placed at the end of the œsophagus or gullet, its object being to more completely triturate the food before it passes into the stomach. The gizzard is composed of hard muscular tissue, and nearly always covered internally with hard growths called teeth.

In Fig. 159, A is the œsophagus or gullet, B the gizzard, D the stomach, E the small intestine, while the large oval-shaped sac at the extreme left is the large intestine.

If the gizzard only is required, the best plan is (after killing) to hold the insect under water, and with the forceps to draw the head from the body. Most of the internal organs will by this means be drawn out, when the gizzard can be detached with the scissors from its place below the gullet, washed, and opened out with knife and needle.

After having proceeded so far, the student will do well to examine the nervous system, and last of all, when the inside has been cleared out, the segments and spiracles may follow in their turn.

Insects intended for dissection should never be allowed to become dry, and if they cannot be treated at once they should be preserved in dilute glycerine, dilute acetic acid, or weak spirit, according to what is required of them afterwards.

The student perhaps may not have patience at first to go through the whole set of dissections from one insect, and, therefore, such objects as the saws of the different saw-flies may be attractive. Several different varieties are shown in Fig. 161.

The variation in the form of saw from the different species, as illustrated in figure, has been carefully studied and the drawings made by Mr. J. W. Gooch. No. 1 is the saw of the large green saw-fly; 2, the black saw-fly; 3, small black saw-fly; 4, a brownish saw-fly; 5, black saw-

fly, with white-banded abdomen ; 6, large yellow saw-fly ; 7, yellow saw-fly with black thorax.

The saw-flies are common everywhere, especially in the neighbourhood of gooseberry bushes ; the saws are really a part of the ovipository arrangement, and are attached of course to the abdomen of the females.

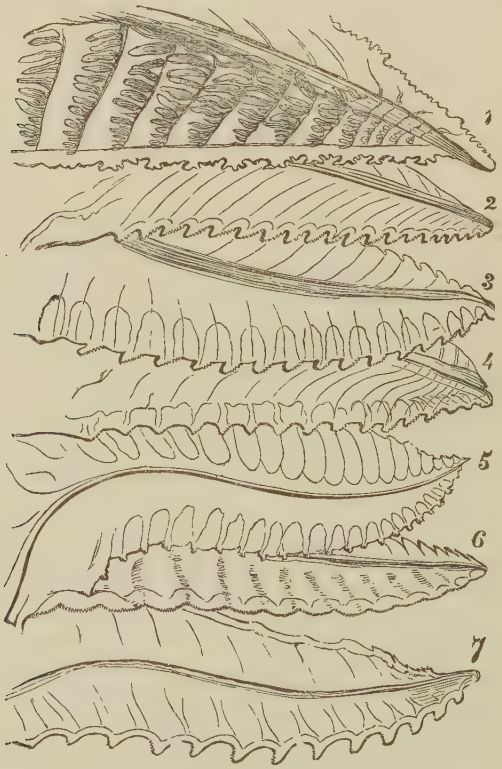


FIG. 161.

The sting and poison-bag of the bee is not difficult to obtain (that of the humble-bee is shown in Fig. 162), though the student will find perhaps that it requires delicate manipulation to separate the long tubular gland from the tracheæ ; this may, however, be done with very smooth

needles under an amplification of about 10 diameters in water and using a small camel's-hair pencil to brush away loose matter.

FIG. 162.

The wasp may also be pressed into the service of the dissector, and here he will find some remarkable differences over the bee; the poison-bag is separated from the body of

the sting by a long tube, and the bag itself is so well covered with muscular fibres that it is impossible to separate them

FIG. 163.

from it without tearing the whole to pieces. The sting, lancet, and poison-bag of wasp may be seen in Fig. 163.

The object of dissecting, after all, is not to be able to prepare pretty slides merely, but to gain a knowledge of the anatomy of our subject, so that we may understand more thoroughly the various transformations which take place. It may not be possible always to isolate each organ we wish, and in that case a careful study should be made of the organs *in situ* sometimes by polarised light, and what we have actually seen may be transferred to paper by any of the methods described in the chapter on "the Delineation of Objects."

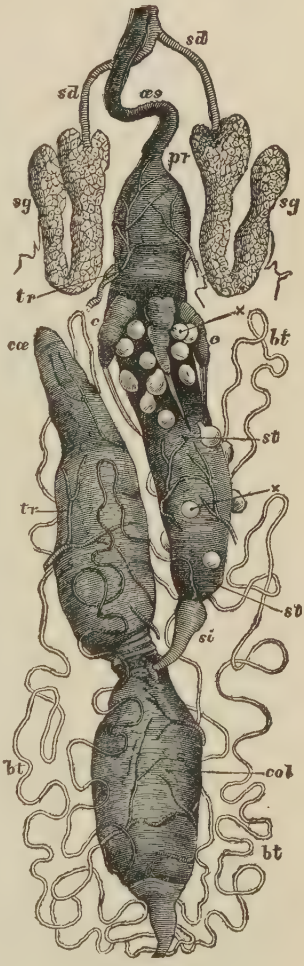


FIG. 164.

An instance of this may be found in Fig. 164, where the internal organisation of the larva of the crane-fly is delineated. To lay out the various organs as there shown would be a task of supreme difficulty; but a drawing may be made of them from the student's observations.

Another study having an interest with many is that of the tongues, palates, lingual ribbons, or odontophores, of molluscs.

The subjects for dissection should be killed by dropping them into glycerine, and so preserved until they are

required. The dissection should be carried on under water, in the usual way, though the mollusc needs but little pinning down.

Let the student start with the common periwinkle, *Littorina littoralis*, or, perhaps better still, with *Patella vulgata*, or common limpet. In the former, the lingual ribbon will be found coiled up like a watch-spring by opening the back of the animal, and this place will be found generally the best of all to examine first.

In *Patella vulgata* it is only necessary to remove the foot, or broad flat disc forming the lower surface of the body, when the lingual ribbon is exposed to view, and appears as shown in Fig. 165.

Fig. 165.

The *Trochus ziziphinus*, or pearly-top, may also form a subject, as it is found in tolerable profusion on our shores; and the whelk, *Buccinum undatum*, will also furnish new specimens.

In *T. ziziphinus*, the floor of the mouth must be exposed from above, when the lingual ribbon will be found lying upon it. In the whelk, the trunk contains the whole of the ribbon, and may be seen by opening the back just behind the tentacles.

In Fig. 166 the periwinkle is well displayed: *f* is the foot, *m* the muscle for withdrawing the animal into his

shell, *g* the spittle glands, *th* the throat, *s* the stomach, *r* the odontophore, *br* the breathing gills, *a* anus, and *o* the ovary carrying eggs.

The next illustration shows an oyster lying in its shell (Fig. 167); *m* is the lower half of the mantle, *m'* a piece of upper half, *g* the breathing gills, *h* the heart, *lv* the liver, *lp* the lips, *o* the opening of the mouth, *a* the anus, *ms* muscle holding shells together, and *c* elastic cushion keeping shells apart.

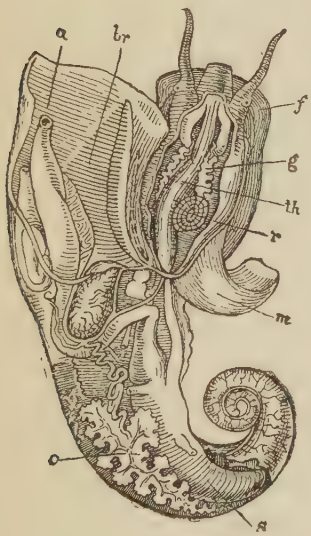


Fig. 166.

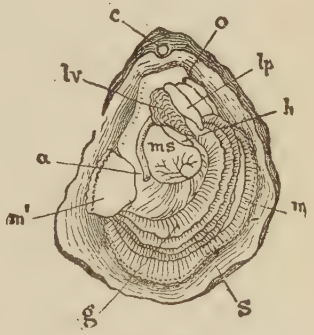


Fig. 167.

Dissection of vegetable matter is usually preceded by prolonged immersion in water, in order to soften the parts to be separated; but very often indeed the tissues withstand this action, and so recourse is had to various other solutions which are found necessary. Caustic soda in dilute solution is a favourite reagent for the differentiation of vegetable matter. It is the solution used by the paper-maker for the disintegration of the leaf of esparto grass, the fibres of which furnish him with a valuable paper-making material.

Fibro-cellular tissue may be conveniently studied in the

leaves of orchids, where it is easily found, underneath the cuticle, after maceration. Woody tissue, often the chief component of certain plants used in the arts, is composed of fibro-vascular vessels aggregated in bundles, as in flax for instance; and the same may be said of jute, china-grass, and many other products. These fibres may be examined by macerating in water, during which a fermentation sets in, and the fibrous bundles consequently split up; the same effect may be produced by boiling with dilute caustic soda solution.

Spiral fibres also form interesting objects for study. They occur sometimes in cells alone, and at others in conjunction with what have been styled bordered pores, as in the yew and araucaria, while the pores are found alone in the pine. Most woods, when in thin shavings, are easily decomposed by boiling with dilute nitric acid, when the fibres can be pulled asunder, or by boiling with caustic soda solution under pressure.

Spiral vessels are even more interesting than any of the foregoing, and easily procured. From the stem of the leek they are separated with ease, while from the petiole of the common garden rhubarb they can be obtained in plenty by searching in a pot of this preserve.

Perhaps of all exercises the complete dissection of a moss is the best for botanical students; the various parts mounted on an ordinary slip under a three-quarter thin glass cover are at once a type slide and an object of beauty.

Scalariform vessels are to be met with in the roots of ferns. The *Pteris aquilina* perhaps displays them as well as any other plant; but it is a moot point whether they are better seen by the examination of a dissected portion, rather than a section made in a diagonal direction. It is true that much can be made out from sections of the

various parts under study; but without dissection we should be very apt to arrive at erroneous conclusions concerning the form and situation of parts, our views often being modified by the light thrown upon our subject by its employment.

CHAPTER VIII.

SECTION-CUTTING.

In the preceding chapter the necessity of dissection has been shown, in order to gain an insight into the economy of things. It is none the less important, however, to be able to cut a thin slice of any given object in order to show the details *in situ*.

We should be very careful not to form hasty opinions, from the observation of sections alone, as vessels will appear of different shapes according to the plane in which they have been cut. Circular vessels cut through obliquely give an oval outline; but in the direction of their length, squares.

Section-cutting may conveniently be divided into three methods:—

1. Without the use of the microtome (section-cutter), and even before hardening or otherwise preparing the subject.

2. With the microtome, such subjects as the stems of plants, leaves, animal preparations, and the like.

3. The cutting of hard substances, coal, rock, bone, and subjects of this nature.

To proceed with the first method, we find some workers cutting slices on or between pieces of cork without the use of any appliance whatever save a knife or razor. Others use even more primitive methods, and perhaps by long continued practice may arrive at fair results, though the author has never seen uniformly thin and good specimens cut by the hand alone.

Some when examining animal tissues, "prefer to snip off

a piece with a pair of sharp scissors," or "cut a piece off with a sharp scalpel," but all slides prepared in this manner are mere fragments compared with the splendid sections of Marsh, Cole, Wheeler, Norman, Ward, and others.

FIG. 168.

Fresh animal tissues, such as kidney or liver, are frequently cut with Valentin's knife (Figs. 168 and 169).

FIG. 169.

This instrument is formed of two narrow blades lying parallel to each other, their distance being regulated by means of a fine screw. The knife is used immersed in water; it is drawn through the tissue to be cut, in a saw-like fashion, the sections being afterwards disengaged by shaking the blades gently in water. Dr. Maddox's form of this knife is one with triple blades, so that a double section might be cut, to show opposite but contiguous surfaces. It is shown in Fig. 170.

FIG. 170.

Dr. Sylvester Marsh, who has written a small manualette upon the subject of section-cutting, has devised a simple but useful spoon for lifting thin sections. It is shown in Fig. 171.

After all, these methods of cutting are exceedingly

limited in their application. They may be necessary at times, when it is desired to make a hurried examination; but the *student* is certainly advised never to make hurried examinations.

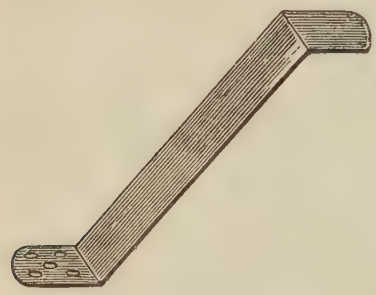


FIG. 171.

We now come to the second division, where sections are cut in the microtome, of which there are several forms, constructed in accordance with the requirements of the originators.

The most common form of section-cutter is shown in Fig. 172. It is generally used for cutting sections of wood, stems, and other semi-hard substances. The substance to

FIG. 172.

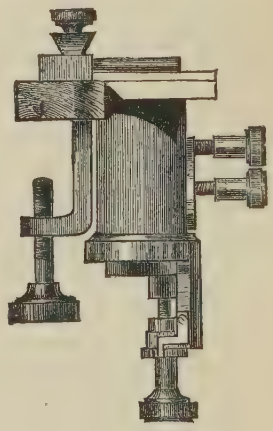


FIG. 173.

be cut is kept firmly in position by means of the side screw, and the quantity necessary for each section sent up by the screw below.

Another section-cutting machine used for the same

purpose as the preceding has been devised by Mr. Hailes, and is sold by Messrs. Baker, of Holborn. All the faults of the old form have been remedied, and for cutting semi-hard substances it is the best of its kind (Fig. 173).

The microtome which the real worker in sections should procure, is that of Professor Rutherford, inasmuch as it has special uses in itself, and can also be made to do whatever

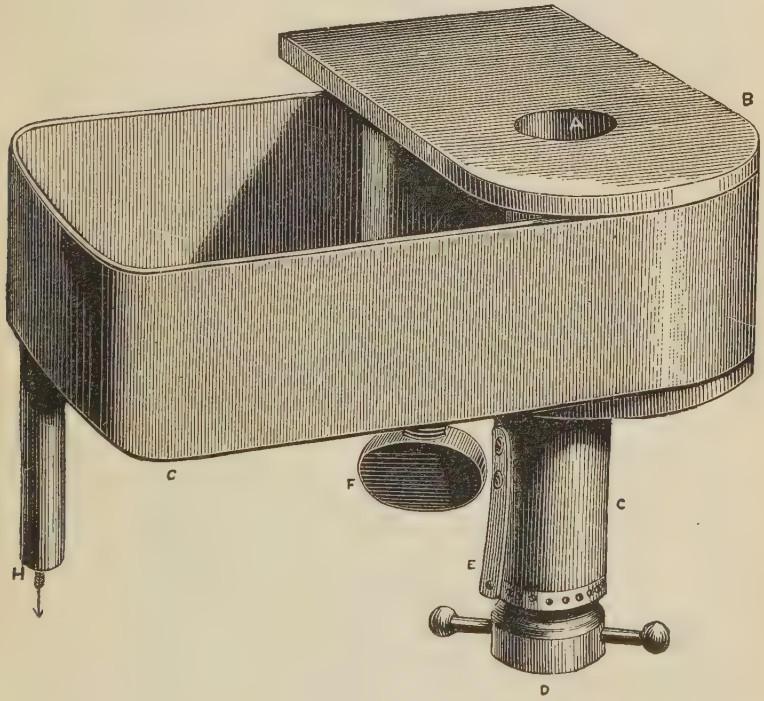


FIG. 174.

has been done with any other machine. It can be used with a cereous bedding or as a freezing microtome at will. The woodcut Fig. 174 shows the instrument.

Mr. Dancer, of Manchester, makes a good microtome for use with a cereous bedding. The well is large and square,

the plug of considerable length, moved upwards by means of a good firm screw, while the cutting plate is of steel. If this microtome were so arranged that it could be used in a freezing mixture, the writer would not hesitate to place it in the first rank, and even before that of Professor Rutherford. The well being square the subject cannot revolve, and no "rise" takes place when the correct bedding agent is employed.

A form of microtome designed by Mr. Williams and made by Messrs. Swift and Son, is shown in Fig. 175. It

FIG. 175.

is specially designed for use in the freezing process, and has been admirably described in the Journal of the Quekett Club. Not having used it the author can but give his opinion that the principle upon which it is constructed is good, though there seems to be very little superiority over Professor Rutherford's.

The student having become furnished with a microtome, his next selection must be a knife or razor, and in his choice he must remember that it is absolutely necessary for the *whole* of the back and edge to lie in the same plane —both must slide together over the top plate with perfect smoothness of motion. The knife or razor should be hollow-ground, flat-ground knives and razors being reserved for cutting wood or bone, where great strength is required. The cutting instruments must be kept to a very keen edge by use of a Washita oil-stone, combined with the application of a good razor-strop. It requires practice to put on the keenest edge. Perhaps a lesson from a barber would on the whole be a cheaper and speedier way of learning.

Mr. M. H. Stiles has described an arrangement by which the blade of the razor is kept from contact with the plate of the machine, by two small screw-clamps he terms "razor-guards" (Fig. 176). These slide on the blade, one being fixed at each end, and kept in position by a small screw in the upper side of each. They are $\frac{3}{8}$ths of an inch wide and are made from sheet brass $\frac{1}{24}$th of an inch thick. They ensure a smoother and more steady motion than by cutting in the old way, and the edge of the razor is preserved in much better condition as it touches nothing but the substance to be cut.

FIG. 176.

Let us now consider the cutting operation. If we wish to cut a section of the stem of the horse-chestnut (*Æsculus hippocastanum*), Fig. 98, say in a line with the axis of growth, we shall find it rather difficult, without first embedding in some agent to hold it firmly, and without undue pressure upon any one part. Formerly a mixture of equal parts of beeswax and olive oil was used for this purpose, and Professor Rutherford advises a mixture of

five parts of paraffin with one part of hog's lard. Both of these mixtures are liable to become loose in the cylinder, often rotating during the cutting operation, and also "rising," and thereby spoiling the work by uneven thickness.

Mr. John Barrow has improved this bedding by making a mixture of naphthalin and stearine in certain proportions, which vary according to the temperature of the air and its consequent behaviour under the knife. It can be mixed in quantity and kept in a large glue-pot, or even a stone-ware jar or preserve-pot, and remelted by standing in hot water when required for use.

This mixture does not become loose in the well of the microtome like the paraffin mixture, and never "rises," so that sections are easily cut of a uniform thickness.

Having soaked the stem in water, in order to soften it somewhat, it should be dried externally and dipped into the following solution, which has been carefully filtered :—

Finest gum-arabic	60 grains.
Glycerine	5 drops.
Alcohol	10 drops.
Water	2 ounces.

The stem must then be withdrawn and allowed to drain upon blotting-paper until surface dry, when it is held in the fluid bedding agent in the well of the microtome, until the naphthalin mixture has become hard, the plug having been previously depressed, by means of the screw, to the depth required.

When quite cold the cutting may be commenced, and the first cut or two made with an ordinary razor and discarded, being only intended to level down and to ascertain if all be right. After levelling, the second cut with the section razor should be examined under a low power, and if it is not thin enough more care must be taken with the

subsequent cuttings. After every cut, the necessary thickness must be sent up by means of the screw below, and if the cutting has been done properly, i. e. by a firm diagonal push from point to handle of the knife, keeping this also back and edge at the same time upon the cutting plate, the section will finally look like the illustration in Fig. 98.

During the cutting operation the knife or razor should be kept flooded with dilute glycerine or weak alcohol, and the section may be easily liberated from the knife by gently shaking in a basin of water. The glycerine and gum surrounding the sections dissolves after a few minutes, allowing them to fall to the bottom, while the naphthalin mixture floats on the surface of the water.

Most vegetable substances can be cut in this way, leaves and stems among the rest; but some will require preparation before inserting into the microtome. Those which are too hard must be soaked in water until soft enough to cut, or even immersed in boiling water if necessary. Excess of water may be abstracted by soaking in alcohol (methylated spirit) for a time, or by partial desiccation. Whenever it is required to eliminate resinous substances methylated spirit will perform the operation, but it must not be forgotten that alcohol extracts the colouring matters from most vegetable substances.

Very good sections of certain woods can be made with an ordinary smoothing plane, such as is used by joiners. Deal may be cut in very thin ribands by this means, and when mounted dry, constitutes Dr. Carpenter's test for the colour correction of objectives.

A word of advice to the beginner may here be necessary: discard all sections not showing the leading features intended; they are worthless from a scientific point of view, yet it is strange how many there are in existence.

Fig. 177 is an illustration of a section of a lichen thallus, and Fig. 178 a similar slice from the apothecium: mounted sections should show the details as well as these.

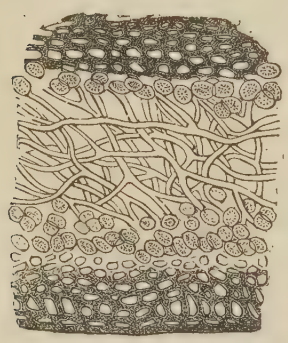


FIG. 177.

FIG. 178.

Animal tissues require different treatment. They are generally too soft and pulpy for immediate cutting, so require to be soaked for various periods in some hardening solution; while others, such as porcupine-quill, whalebone, and horn, require immersing in hot water to soften them.

For hardening animal tissues, alcohol, and chromic acid solution are most generally used, while some prefer bichromate of potash, or bichloride of mercury. Alcohol is perhaps the safest of all hardening agents; it being a powerful abstractor of water, coagulating albumen, and acting also as a preservative agent.

Chromic acid is to be obtained in beautiful dry carmine crystals. It is more conveniently kept as a stock solution, 1 ounce being dissolved in 50 ounces of water, and diluted as occasion requires.

It must not be forgotten that tissues are easily over-hardened and rendered brittle by prolonged immersion in chromic acid, and therefore the weaker a solution can be used the better. Some tissues require hardening by special

means. Rutherford advises brain to be soaked in the following solution :—

Chromic acid	15 grains.
Bichromate of potash	31 grains.
Water	43 ounces.

Small pieces are first immersed for twenty-four hours in methylated spirit, drained on blotting-paper, and then soaked for five or six weeks in a large quantity of the above solution, changing it several times in that period. Adipose tissue may be hardened in methylated spirit; liver prepared for cutting by soaking in alcohol, commencing with weak methylated spirit and finishing in absolute alcohol; lung in chromic acid, muscle in chromic acid, as well as tongue, stomach, and spinal cord—the last in one part of the stock solution diluted with nine of water, as it is extremely liable to over-hardening. The actual operation of hardening may be performed in the following manner: Cut the substance—kidney, for instance—into pieces half an inch square, and about the length suitable for the well of the microtome. Place these in methylated spirit diluted with an equal bulk of water for three days, drain well upon bibulous paper, and then immerse in a solution of chromic acid prepared by diluting one part of the stock solution with seven parts of water. Allow the pieces to remain in this for three days, then pour it off and replace it by fresh. At the end of ten days a piece may be abstracted and a rough section cut in order to see whether it has become sufficiently hardened; if not, the chromic acid must be again poured off and the pieces covered with fresh solution, making an examination of it with the razor every three days. Always harden insufficiently in the chromic acid, and when just under that degree required, take it out and put into methylated spirit to cleanse.

Pour off the spirit every day and replace it with fresh until the excess of chromic acid has been abstracted, known by the spirit remaining clear and colourless.

Injected kidney, and, indeed, most other *injected* tissues, must be hardened entirely in alcohol, as the chromic acid, being a powerful oxidiser, would in all probability cause a decomposition of the colouring matter. Kidney is often injected with Prussian blue. In this case it is as well to add a few drops of hydrochloric acid to the alcohol used in hardening.

The hardened tissue is now ready for cutting, and this operation may be illustrated by employing the Rutherford microtome, illustrated in Fig. 174.

The substance when taken out of the spirit must be placed on blotting paper, and allowed to become surface dry. When this is effected it must be dipped into the gum solution before mentioned, and, after removal, is to be again laid upon the paper until dry.

The microtome having been screwed to the table, and the plug lowered by means of the screw, the melted mixture of naphthalin and stearine is to be poured into the well. When just about to set, the substance is inserted carefully, and held in position until the bedding has sufficiently solidified. After an hour or more, according to the temperature of the atmosphere, the substance will be in a fit state for cutting, and may be performed in exactly the same manner as described for vegetable stems.

The section knife is best kept in order on an ordinary razor-strop, with an occasional touch on a fine Washita oil-stone.

Fig. 179 shows a longitudinal section of the injected kidney of a rat, *a* being the arterial trunk, *b* the venous trunk, and *c* the glomerulus ; while Fig. 180 is a section of human muscle showing *Trichina spiralis in situ.*

Rutherford's microtome is also capable of being used as a freezing instrument, in a manner easily understood by reference to the accompanying illustration, it being a section of the instrument already figured.

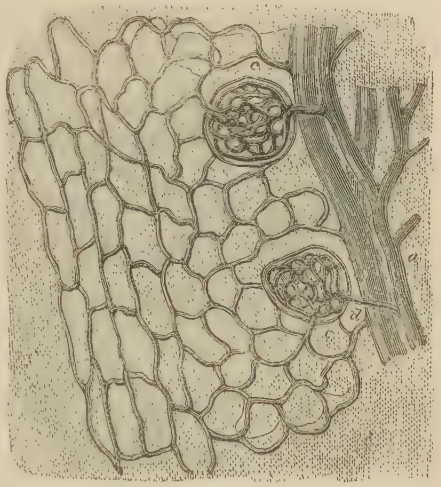


FIG. 179.

The freezing box is surrounded with several layers of flannel, and the machine screwed to the table. The plug K being unscrewed, pour methylated spirit into the tube C, oil the sides of the plug, replace it, and depress the screw to the desired extent. Remove all spirit which has come above the plug K, and carefully close the margin with some hog's-lard to prevent the spirit reaching the cavity above the plug.

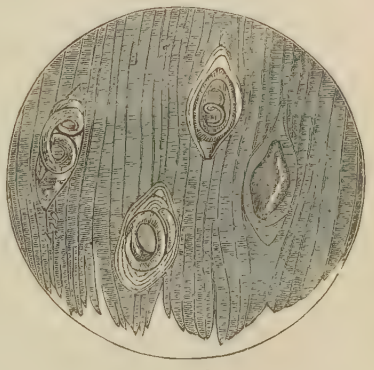


FIG. 180.

The embedding fluid is made by soaking five ounces of picked gum arabic in ten ounces of water, and when it has dissolved add two drachms of camphorated spirit and one drachm of glycerine, after which it is to be strained through calico, and kept in a bottle, free from dust. This mixture, when frozen, cuts very much like a Gruyère cheese.

In working, place in the freezing box alternate layers of finely powdered ice and common salt, taking care that they are pushed well round the tube of the machine; attach an indiarubber tube to H and conduct the water into a jar or pail, seeing also that it is kept open in order that the liquid may find easy egress. The mixture of ice and salt must be added at intervals, and the contents of the box stirred occasionally with a short rod in order to promote the escape of fluid.

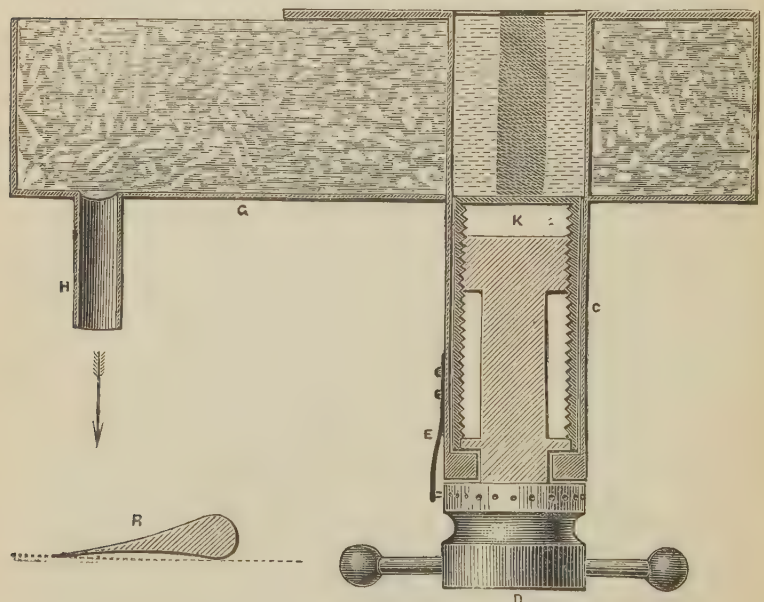


FIG. 181.

The gum solution is run into the well of the microtome and covered with a piece of guttapercha; when the film of ice has formed at its periphery, the tissue should be held against the advancing film until it becomes fixed in position for easy section. The razor used should be of the form already described (R, Fig. 181), and when in

use with the freezing microtome it will be unnecessary to wet it.

The top plate would be better made of steel, or even of glass, unless razor-guards be used, as though it is easy after some practice to prevent cutting into the brass, yet this invariably happens to beginners. Do not forget to *well* wash the microtome and knife before putting them away, and to grease them with some heavy lubricating oil.

We now come to the preparation of sections of hard substances, such as bone, coal, rocks, and minerals generally. Some operators start with a fragment chipped from a large specimen; but without doubt when several sections have to be cut, the best plan is to resort to what is termed slitting, being done by the microscopist himself or sent to the lapidary, who will cut slices for a very small charge.

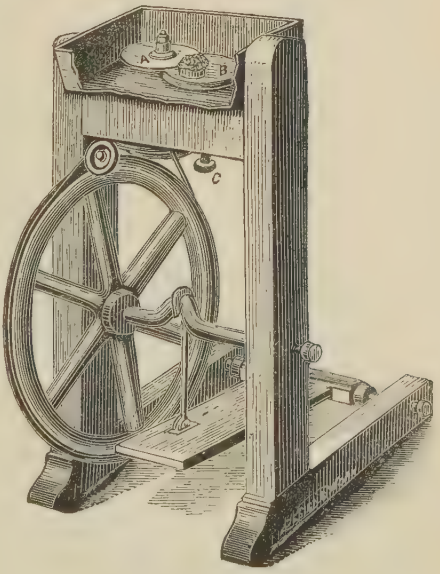


FIG. 182.

If the operator desires a bench of his own, it would be advisable to have the slitter and laps interchangeable, so that they may run on the same centres. The woodcut, Fig. 182, will show how such a bench may be constructed.

The slitter is a thin wrought-iron disc about 11 inches in diameter by about $\frac{1}{50}$ of an inch in thickness, and when used

its edge is charged with diamond dust in the following manner: Reduce a small splinter of diamond to a fine powder on a hard steel plate, then run a small quantity of tallow on to it, and mix thoroughly with the steel crusher. Press the tip of the longest finger into the mixture, bringing away a thin coat. The slitter should then be moistened with petroline, and a bloodstone or agate pressed gently against it; at the same time the finger should be brought over the revolving disc in such a position that the edge may just scrape the tallow off. This should be evenly distributed round the edge of the disc, which is readily done by rapidly touching it while revolving.

The rock, fossil wood, or other mineral, is now to be ground flat on one side, and firmly cemented by old balsam or marine glue (solid) to a glass slide, say 3 × 2 inches and $\frac{1}{4}$ inch thick. This can readily be slid along the guide-plate B, insuring perfect parallelism, the requisite thickness of each section being regulated by the screw C, which raises and depresses the guide-plate. The sections may be about $\frac{1}{16}$ of an inch in thickness, more or less, according to the subject, but the thinner they are cut, so much more labour is saved in the subsequent operations. The sound indicates when the slitter is cutting properly.

One side must be ground down on the leaden lap, which may be substituted for the slitting disc A in the bench shown in Fig. 182. This lap is about 10 inches in diameter and $\frac{3}{4}$ of an inch thick; it is used with fine emery and water for the first grinding, the final being performed with still finer emery upon a ground brass lap, preferably made to run in the same direction as the face-plate of a lathe—in fact, one of the cheap lathes shown in Fig. 183 will do exceedingly well for this work, and no doubt the ingenious student will be able to rig up a slitter and leaden lap to fit this form of lathe,

which may be made a very useful tool by the practical microscopist.

In the grinding operation the student must remember that a *polish* is not necessary. The Canada balsam in which the section is mounted produces an apparent polish.

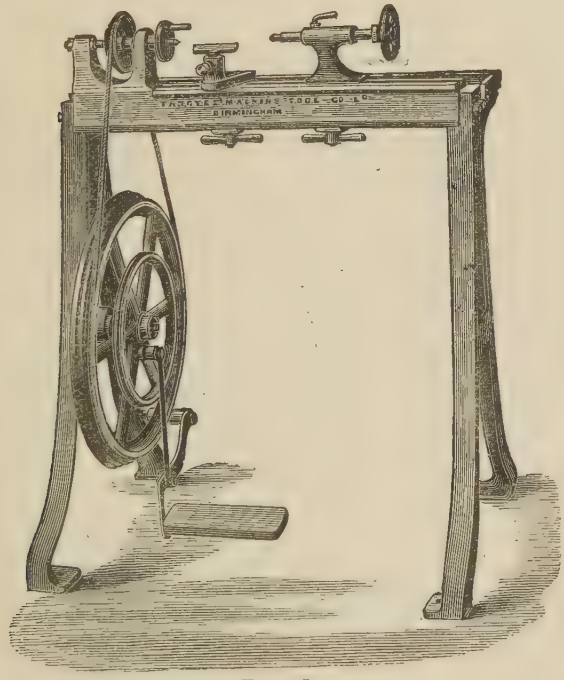


FIG. 183.

All that is required is to produce a very smooth surface free from scratches, and this is readily done with a brass lap and the finest emery procurable. The one side of the specimen having been brought into the above condition, is to be cleaned and firmly cemented with old balsam to the centre of a 3 × 1-inch glass slide. The balsam is dropped upon the centre and warmed over a lamp until, when cold, it is just possible to produce an impression with the nail.

The balsam is then to be rendered fluid again, and the polished section made gradually hot, when it is let down gently, taking care to avoid air bubbles, pressed down firmly and then put aside to set thoroughly. After several days, the rough side may be ground down on the leaden lap with emery, and finished off on the brass lap with the finest emery. The slide is now finished off by cleaning with spirits of wine and a camel's-hair brush, then dropping some fluid balsam upon the centre of the section and covering in the usual way with thin glass. If the glass of the 3 × 1-inch slide be disfigured with scratches it may be covered with an ornamental cover-paper, sold by Wheeler and other dealers in microscopic specialities. The foregoing method will produce good results for almost any kind of work; but of course there are simpler methods which may suit the student, requiring however much more labour. One of our friends grinds down roughly upon a grindstone or emery wheel, and finishes upon an Arkansas oil-stone. It is rather tedious work, but produces good results.

Mr. J. H. Jennings gives the following simple instructions:—

1. Preparing sections of hard rocks: In the first place a thin chip must be procured by the use of a hammer. This chip should be about 1 inch square, and not more than $\frac{1}{8}$ inch thick; chips of sedimentary rocks may be thicker. Rub the chip down by hand with emery and water on an iron plate, until one side is perfectly flat. To remove scratches, next rub the chip on a glass plate with fine emery, and polish on a Water-of-Ayr stone; when quite smooth, wash it well and let it dry. Meantime place some old balsam on a glass slip, and warm it over a lamp until all the more volatile parts of the balsam evaporate, so that on cooling it becomes hard and tough. Do not let the

balsam boil. When the balsam is properly hard, warm the chip gently over the lamp or on a hot metal plate, brush it over with a little turpentine, and re-melt the balsam; then lower the chip slowly into the balsam until it is cemented firmly and evenly by its flat surface to the glass slip. When the balsam is quite cold, the chip is to be rubbed down on the iron plate with coarse emery until it is too thin to bear any further rough friction. With care, many rocks may be brought to the requisite thinness on the iron plate alone, and will require little finishing. The necessary degree of thinness will vary according to the nature of the rock; but, as a general rule, most hard rocks must be cut thin enough to read through when placed on the page of a book. When the section will no longer bear the friction of the coarse emery, remove it to the glass plate, and grind it thinner with flour emery, and finally finish it off on the Water-of-Ayr stone. The slide, at the finish, will be disfigured by deep scratches from the emery, and the section must be transferred to a clean slip. Warm the section enough to melt the balsam, and push the section off with a needle into a cup of turpentine, and wash carefully with a small brush. Now pour a little balsam and benzol solution on the clean slip, place the section upon it, add a little more balsam, and cover.

2. Preparation of soft rocks and sedimentary rocks generally: These are prepared and mounted in the same way as hard rocks, *but no emery is to be used;* they must be ground down and finished on the three stones mentioned above. Some very friable rocks will require a preliminary hardening by immersion for some days in a solution of (1) balsam in benzol. The balsam must be first baked in a cool oven, until on cooling it becomes hard and brittle; then dissolve it in benzol; or (2) in a solution of shellac in alcohol. This is, perhaps, the better of the two. When

the chip has remained long enough in the solution it must be dried in a warm place. Sedimentary rocks, as a rule, do not require to be cut as thin as igneous rocks, so that they may be left on the grinding slide, as it will not be scratched.

One thing is absolutely necessary in either method, that is to get rid of *all* air bubbles in the balsam attachment, as if any are left, the section is sure to wear into holes and break.

Sections of Echinus spine may be cut in the same way as above, or with a fine saw improvised from a thin clock spring, and the slice ground down by rubbing on a fine level Turkey hone, and when thin enough should be cleansed with water and a soft camel-hair brush, dried by immersion in alcohol, passed through benzol, and finely mounted in balsam and benzol, or dry, if required, when it is ready for observation.

Hard rocks, as a rule, are easier to prepare than soft ones. The latter should be soaked in turpentine, and then in balsam and benzol, afterwards being heated till quite hard.

Coal may be cut and ground into sections in the following manner:—After careful selection of a piece free from cracks, a prism is to be cut $\frac{1}{2}$ inch or $\frac{3}{4}$ of an inch square. Slices of this, as thin as possible, may now be cut with a fine saw, and one face rubbed flat upon a slab of pumice-stone kept well wetted with water, and finally rubbed smooth upon a Water-of-Ayr stone, water being continually applied to it. The polished surface is now cemented to a 3×1 glass slide with marine glue, taking special care not to include air bubbles, and the slide put by to set for some days. When this has taken place, the slice may be rubbed down on a flat piece of coarse gritstone with plenty of water until almost thin enough to show the structure, and

when arrived at this stage, finished off first on the pumice slab with water, and finally on the Water-of-Ayr stone.

The study of coal sections gives us much insight into the formation of that peculiar substance. Fossil fungi have been found in coal, and described by Mr. Worthington Smith as belonging to the genus Peronospora, and the plants have therefore been called Peronosporites. They are shown in Fig. 184.

Bone and teeth are generally cut with a saw, and afterwards rubbed down with a hone and water. Dr. Beale

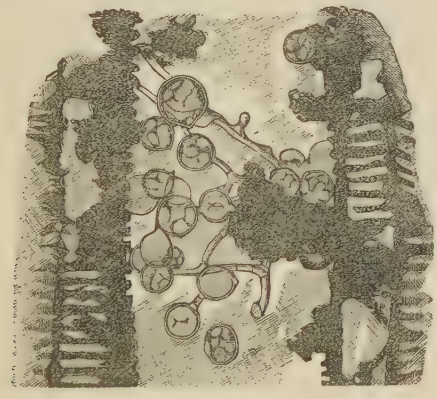


FIG. 184.

objects to this method, as filling up the lacunæ and canaliculi with debris, and advises cutting a thin section with a sharp knife, afterwards staining with carmine. Mr. George Hoggan described a machine for cutting sections of hard substances to the members of the Quekett Club, in which he stated that each section was cut in two or three minutes, which, after brushing away the sawdust, was ready for mounting in balsam.

A few months since, Dr. Matthews described a machine for a similar purpose at a meeting of the Royal Microscopical Society, an illustration of which is shown in

Fig. 185. Such a machine is clearly only intended for specialists, and therefore a full description of it would be out of place here.

Dissections and section-cutting are generally looked upon by the student with awe, as being exceptionally difficult. This is not the case: with proper care and perseverance almost any one may become an expert dissector or section-cutter, and it is to be hoped that whoever purchases a microscope through reading these pages will endeavour

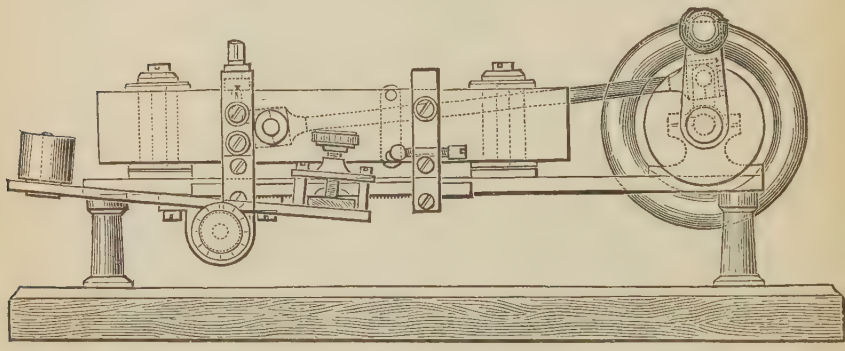


Fig. 185.

to do some useful work, and not be content with mounting a few crystals for the polariscope, or with soaking an insect in potash, and mounting in balsam, believing that is all that need be done with it.

CHAPTER IX.

THE DELINEATION OF OBJECTS—MICROSCOPIC MEASUREMENTS.

We have now to dilate upon the importance of sketching everything of interest the observer may see under the microscope.

Dr. Beale says: "The student cannot too soon try to delineate what he demonstrates. He will teach himself to observe the more accurately and the more quickly if he record the results of his work in pencil sketches. A truthful drawing of what a man has recently seen may be compared with drawings made 100 years hence; and although the means of observation will be more perfect than they are at present, such comparisons may be useful in many ways, and especially in preventing erroneous conclusions from becoming popular."

It cannot be too strongly impressed upon the reader, of the advisability of frequent practice with the various means devised for this purpose. Microscopical contributions to our scientific papers, communications to societies, and even the results of our every-day observations, all become more interesting if illustrated in some way or other.

There are many methods by which microscopical objects may be delineated. They may be roughly drawn by means of pencil and paper, guided by the eye alone or assisted with the use of a circle of glass ruled into squares, made to rest upon the diaphragm of the eye-piece; the paper, in

this case, may also be faintly ruled into squares, and by this means, especially after a little practice, very accurate drawings can be made. Enlargements and reductions can also be just as easily drawn. A glance at Figs. 186 and 187 will illustrate this. The former represents what the observer is supposed to see upon looking down the eye-piece of the microscope; while the latter delineates the figure produced by the pencil upon the ruled paper.

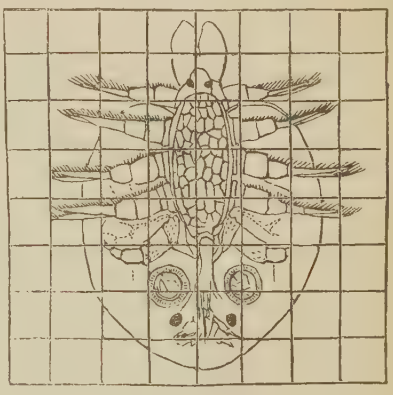


FIG. 186.

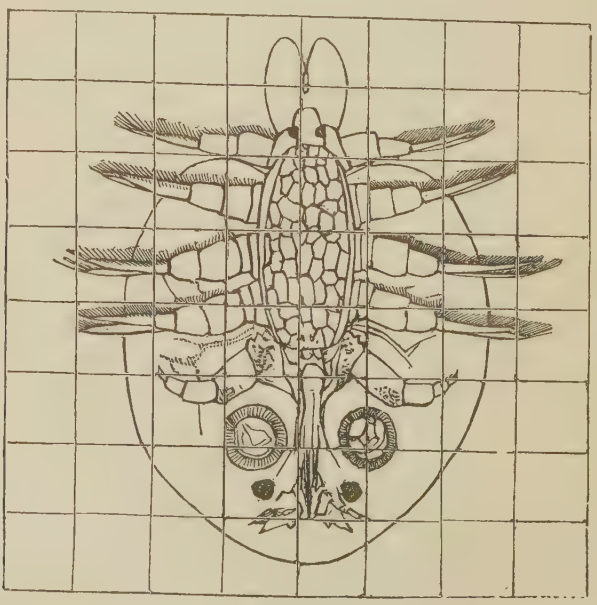


FIG. 187.

Another method for the delineation of objects is by the use of the camera-lucida or neutral tint reflector, which by a little practice becomes a very accurate process. The ordinary camera-lucida consists of a prism, and apparently throws down an image of the object upon the paper below it.

This is the same with the neutral tint reflector; it is made of a small piece of plate glass, slightly coloured and arranged at an angle of 45° with the eye-piece; it is shown in Fig. 188.

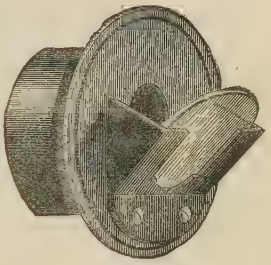


FIG. 188.

There is a fault with nearly all reflectors hitherto made; the glass is too thick, and consequently two or more images are formed of the object, which come into view by a slight movement of the head. To remedy this, a writer in 'Science-Gossip' proposed a head-rest, but even this may be dispensed with if the glass used be of the same thickness as an ordinary cover, such as is used in the mounting of objects. In the construction of a reflector for drawing purposes, it is only necessary to hold a thin glass cover at an angle of 45° with the eye-glass, in such a position that the centre of the cover coincides with the optical axis of the microscope. Surely students interested may manage to do this for themselves.

When using either the camera-lucida or the neutral tint reflector, the cap is removed from the eye-piece and the accessory placed in its stead, the microscope arranged horizontally, and the paper placed under it upon the table, as shown in Fig. 189.

The instrument shown is one of Messrs. Ross's make, and is nicely balanced in all positions. Messrs. Swift and Son place a stop on most of their stands, so that it is easily known when the body is either horizontal or vertical.

The most important point to be remembered in the use of these reflectors is the proper management of the light. Perhaps the student will fail at first in seeing *together* the pencil and the object, but this difficulty will vanish upon securing the proper illumination. The light must not be concentrated too strongly upon the object, neither should the paper be placed entirely in the shade, the student soon arriving at the happy medium after a few trials.

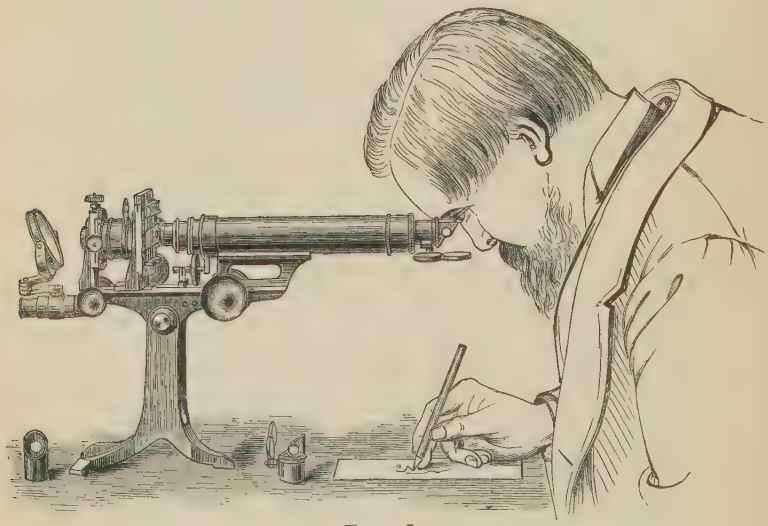


FIG. 189.

In order to obtain the same degree of amplification upon the paper as appears when looking down the tube of the instrument, the paper must be placed at the same distance from the camera-lucida as that accessory is situated from the objective front; but the magnification is generally expressed at a distance of 10 inches.

Another form of this instrument is Nachet's, for use with vertical microscopes, with the ordinary pattern when used vertically with immersion lenses or in any other position of the instrument.

A diagram of this form is shown in Fig. 190, from which the reader will see that it consists of a prism of nearly rhomboidal form placed with one of its inclined sides over the eye-piece of the microscope. To this is cemented a segment of a small glass cylinder, constituting the complete apparatus. This enables sketches to be made with the microscope body disposed vertically with the greatest ease, as although the ordinary camera-lucida or neutral tint reflector can be used with the microscope in this position, still drawing from it in this way is not a comfortable operation.

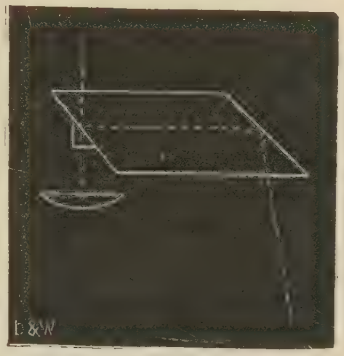


FIG. 190.

A camera-lucida somewhat similar to the above is made by Mr. Swift and shown in Fig. 191 ; it can also be used at any inclination of the optical tube of the microscope.

There is another method by means of which microscopic objects may be faithfully delineated, and that is by photo-micrography. This art is usually considered a difficult one, and when we are confronted with the fact that nearly all who have written anything upon the subject have advised the use of a host of complicated paraphernalia, it is scarcely surprising that such an opinion should have gained ground. The vast array of apparatus, condensers of peculiar construction, blue cells, heliostats, and even to a special room set apart for the camera alone, must have been quite sufficient to frighten any one with only their evenings at leisure.

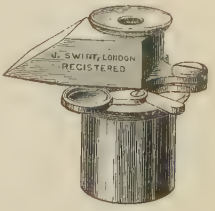


FIG. 191.

An ordinary microscope and photographic camera, as shown in Fig. 193, is all that is absolutely required for this work, though with some it may be more convenient to use the apparatus illustrated by Figs. 192 or 195.

The camera proper is made to remove from the base-board, but this is for portability only. The base-board should project for some distance in front of the camera to carry the microscope and illuminating apparatus as in Fig. 193, so that the whole may form a solid continuous base. This is necessary, as well as a good firm table upon which to place the apparatus, and this must be quite free from any vibration to insure perfect sharpness of the image.

Of course the length of the base-board and of the camera body when extended must vary with the amplification required. The diameters to which an object is enlarged in the camera (Fig. 192) when the sensitive plate is at a distance of 36 inches from the object, may serve as a guide in this direction.

Designation.	Diameters.	
	Without Eye-piece.	With the A Eye-piece.
4 inch	12	36
2 ,, ...	21	63
1 ,, ...	37	110
$\frac{1}{2}$,, ...	80	240
$\frac{1}{4}$,, ...	173	520
$\frac{1}{8}$,, ...	360	1000
$\frac{1}{16}$,, ...	530	1600

If the operator prefers to work with the eye-piece in addition to the objective, the camera need only be a short one, say extending from four inches to twelve, the apparatus being arranged as shown in Fig. 193. At this latter distance he will get the same degree of amplification

as when looking down the tubes of his microscope, and the author has but few doubts whether, after all, despite adverse criticisms, this is not the simplest and cheapest plan. He

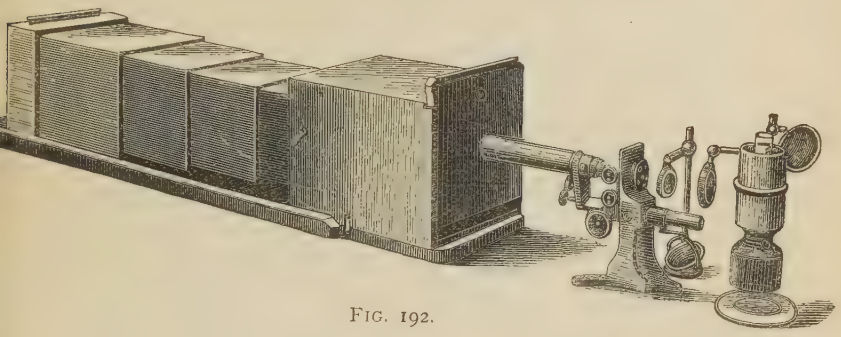


FIG. 192.

has both taken, and seen taken, photo-micrographs of the same subjects, with and without the use of eye-pieces, and could not discover any difference between them.

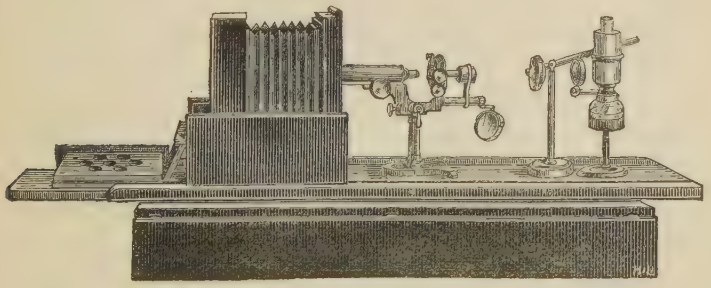


FIG. 193.

There is more light lost when working with the eye-pieces attached, but in these days of extra-sensitive plates this is certainly no disadvantage with the lower and medium powers.

The microscope employed may be of any ordinary kind, monocular preferably, and if provided with a means of shortening the tube or body so much the better. The tube should be lined with velvet, to prevent central flare,

and the instrument provided with a coarse and fine adjustment, the latter answering the helm when turned either backwards or forwards; but more particularly must it be sensitive to very slight movements of the milled head when *withdrawing* the objective from the stage.

Dr. Woodward states "the objective selected should always be specially corrected for photography." The author is not able to agree with Dr. Woodward on this point; no doubt it is very convenient to have one so corrected, but it is by no means imperative. To those as yet uninitiated in the mysteries of the photographic art, it may be necessary to say that the *visual* and *actinic* foci often (though not necessarily) lie in different planes, and therefore an objective which gives perfect definition *to the eye* will often produce a blurred and indistinct image upon the sensitive plate. This may be remedied in withdrawing the objective, by turning the milled head of the fine adjustment; it cannot be told beforehand what allowance is necessary, the only method is by trial and error; some objectives require no correction whatever, while others which necessitate considerable movement from the object, can be made to yield just as perfect results.

The best way to arrive at the necessary correction is to take a picture at the best *visual* focus, develop, fix, and dry in the usual manner, and then withdraw the objective from the stage by means of the fine adjustment until the image appears on the ground glass about as indistinct as it does in the negative. This will probably be the amount of correction required, and one or two trial plates will decide the question. For powers up to the half-inch the proboscis of the fly (Fig. 99) is a good object to experiment with; a photograph of this preparation, taken with a Ross's 1½-inch, showed that its actinic focus was identical with

its best visual, while a 2-inch, by Dancer, required one complete revolution of the milled head of the fine adjustment.

Ordinary ground glass is too coarse to focus upon; two surfaces should be rubbed together with a little of the finest emery and water, or a focussing glass may be prepared by coating an ordinary glass plate with the following varnish:—

Gum mastic	40 grains
Gum sandarac	160 grains
Ether	4 oz.
Benzol	1½ oz.

The correct focus may be obtained by the eye alone, though this is difficult, especially with high powers; and, therefore, it is advisable to employ a magnifier of some description—an engraver's lens or an ordinary eye-piece with the eye-glass removed as shown in Fig. 194. The field lens is placed next the eye and the open brass tube slid in a parallel direction over the glass focussing screen.

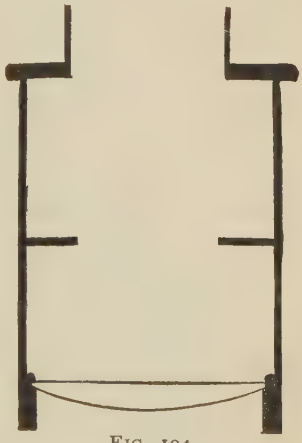


FIG. 194.

The author now uses a much improved method for focussing:—removing the ground glass slide another is substituted of mahogany, but pierced with a series of seven holes, into each of which the ordinary A eye-piece may be fixed. The thickness of the slide is such that when the eye-piece is pushed in as far as it will go the diaphragm lies in the same plane as the ground surface of the

glass slide. To any one accustomed to focus by the old method the present system will be found a considerable improvement, it being *easy* under these conditions to obtain a sharp focus with an ordinary paraffin lamp when using the $\frac{1}{16}$ objective. (See Fig. 193.)

The microscope stand employed by the author possesses a fine adjustment, moving the objective $\frac{1}{200}$ of an inch for each revolution of the milled head, and is divided into twenty parts. His objectives require the following corrections :—

Designation.	Maker.	Air Angle.	Correction.
4-inch	Browning ..	9°	0·0100 inch
2 ,, ...	Dancer	14°	0·0050 ,,
1 ,, ...	Browning ..	25°	0·0025 ,,
$\frac{1}{2}$,, ...	Browning ..	60°	0·0005 ,,
$\frac{1}{4}$,, ...	Browning ..	85°	0
$\frac{1}{8}$,, ...	Browning ..	140	0
$\frac{1}{15}$,, ...	Dancer	170°	0

The source of illumination may be the ordinary microscopical paraffin lamp, as the light from it is much more actinic than that of gas, and for all powers below the $\frac{1}{4}$-inch it is only necessary to use the bull's-eye condensers as shown in the figures.

A very intense illumination, for photo-micrography, may be obtained from paraffin oil by using it in a triple-wick lamp, such as the Triplexicon or Sciopticon, with a condenser parallelising the rays proceeding from it, as shown in Fig. 195. These parallel rays are again to be converged by a convex lens of 3 inches diameter and 10 inches focus. Such an arrangement as used by the author is shown by Fig. 195, and by means of it the relative lengths of exposure have been very much shortened.

With the objectives already alluded to, the sensitive plate being at a distance of 24 inches from the object, with

an ordinary paraffin lamp and bull's-eye condenser of 3 inches focus, the shortest exposures for good pictures

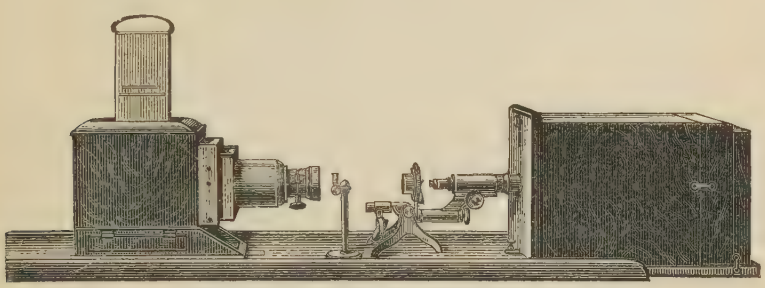


FIG. 195.

with Mawson and Swan's "15 times" sensitive plates have been found as follows:—

Designation of Objective.	Subject.	Exposures.	
		By Method Fig. 192.	By Method Fig. 195.
4-inch ..	Wing of blow-fly	6 seconds	3 seconds
2 ,, ..	Proboscis of do.	60 ,,	10 ,,
1 ,, ..	Do. do.	70 ,,	20 ,,
$\frac{1}{2}$,, ..	Glass crystal	60 ,,	30 ,,
$\frac{1}{4}$,, ..	Section of deal	7 minutes	2 minutes
$\frac{1}{8}$,, ..	Podura scale	10 ,,	$2\frac{1}{2}$,,
$\frac{1}{16}$,, ..	*Pleurosigma attenuatum*	15 ,,	3 ,,

Dr. Carl Seiler, in a communication to the 'American Journal of Microscopy' upon this subject, seems to prefer wet plates to dry ones, and adduces arguments in support of his views, one of which is cheapness. The cost of plates by the wet or dry process is so small that this item need not enter into the calculation; moreover, dry plates are rapid, so convenient, and with good manipulation give such excellent results, that the author pleads for the use of them exclusively in producing negatives.

There are many kinds of dry plates to be met with in the market, nearly all of which are now prepared from gelatine

emulsion. Mawson and Swan's, having been extensively used by the author for this kind of work, deserve commendation as being of good and uniform quality.

Do not over-expose these gelatine plates, or you will get nothing but thin and misty images. From some cause, those exposed to gas or lamp-light do not *commence* to develop so quickly as those exposed to sunlight; but if time is given, and they are not over-exposed, perfect pictures may easily be obtained. The time of exposure in the camera depends upon the quality of the light, *but more perhaps upon the nature of the object than anything else;* for instance, a section of the kidney of a horse required twenty minutes' exposure with the argand gas and bull's-eye condenser, while a crystal of glass under the same conditions of light and amplification required but one minute.

For ascertaining the exact exposures and the corrections required for the difference in actinic and visual foci, the beginner is strongly advised to keep a bound record of his failures as well as prints from *all* his negatives, with full particulars respecting them; they are great helps in photographing new subjects.

Mr. Dancer of Manchester, Dr. Maddox, Mr. Shadbolt, the late Dr. Redmayne of Bolton, Mr. Wenham, and several others have been the most successful photographers of microscopic objects; not forgetting, however, Colonel Dr. Woodward in America, as most of the photographs and nearly the whole of the literature of this country has emanated from him.

Dr. Woodward has described his method with sunlight and a heliostat, from which we gather the following:—

The microscope being placed on a shelf at the window of the dark room, and its body made horizontal, the achromatic condenser is illuminated by a solar pencil reflected from a heliostat (Fig. 196) upon a movable mirror outside

the shutter, and thence into the dark room. No ground glass is used, but a lens mounted in a suitable tube is fixed in the opening of the shutter through which the solar pencil enters. This lens is an achromatic combination about 2 inches in transverse diameter and of about 10 inches focal length. It is placed at such a distance from the achromatic condenser that the solar rays are brought to a focus *and begin again to diverge* before they reach the lowest glass of the achromatic condenser.

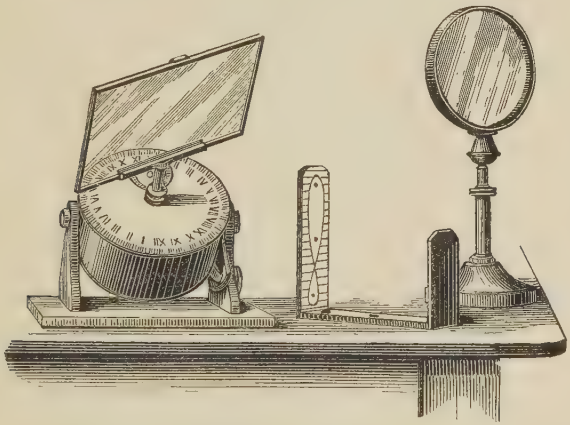


FIG. 196.

All trouble from the solar heat is thus completely avoided. So successfully may this be done, that it is possible to obtain light enough to give distinct vision and admirable definition on the cardboard screen with 5000 diameters, while the heat is so slight that the drop of water used with the immersion lens does not require renewal oftener than about once in two hours.

For anatomical preparations requiring for their display from 200 to 500 diameters, the $\frac{1}{8}$-inch objective may be used without an eye-piece, obtaining the precise power desired by variations in the distance of the sensitive plate

from the stage of the instrument. Dr. Woodward gives the preference to immersion objectives, the corrections of which are generally well suited to photographic requirements.

With a $\frac{1}{8}$ objective and the arrangement above described, the field is so brilliantly illuminated that the eye cannot safely be permitted to look down the tube. The image is therefore received on a piece of white cardboard, and, sitting by the microscope to make the adjustment, the card is viewed with both eyes, precisely as in the case of the ordinary solar microscope.

When all is satisfactory, an ammonio-sulphate cell is inserted between the large lens and the achromatic condenser, and the velvet hood, which prevents leakage of light from about the microscope into the dark room, is drawn down.

Dr. Woodward finds it best to use the naked objective without eye-piece or amplifier, and not as a rule to fix the sensitive plate more than 3 or 4 feet from the stage of the microscope. A $\frac{1}{8}$ objective may be conveniently employed to obtain powers of from 200 to 500 or 600 diameters; a $\frac{1}{16}$ for higher powers, up to 1200 or 1500 diameters.

It occurred to him that for such short exposure the heliostat might be dispensed with, and on trial without it found that a large right-angle prism used in the position of total reflection, or even an ordinary mirror, gave excellent results; the exposures being even shorter than when the heliostat was used, since there was but a single reflection. Under ordinary circumstances the heliostat appears desirable for general use since, the solar pencil being thrown in a constant direction, the trouble of adjusting the illumination of a series of objects is considerably diminished; but equally good pictures can be produced without it, even

with very high powers, a circumstance of considerable interest where motives of economy preclude the microscopist from procuring this convenient instrument.

The time of exposure required for the production of pictures magnified 500 diameters or less was about a second. With higher powers it increases, varying with the management of the achromatic condenser. For 4000 diameters as much as twenty-five seconds is sometimes required.

In using the electric light it is necessary to render the divergent pencil proceeding from the carbon points as nearly parallel as possible, by means of the condenser usually supplied with electric lamps for this purpose, and then introduce into the parallel pencil, instead of a ground glass, the very same condensing lens described above for the process with solar light. The image is received primarily for focussing on a cardboard screen, and the

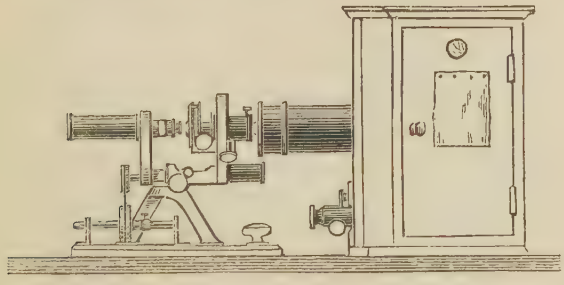


FIG. 197.

remaining details do not differ from what has been related above. The time of exposure does not exceed a single second for 400 diameters, and the sharpness of the pictures is excellent. For well-made tissue preparations, however, the best work obtainable with the electric light is so similar to the best attainable by sunlight, used as above described, that it is scarcely necessary to take the trouble

to set up the battery and work the electric lamp, unless it was desirable to work at night or in unfavourable weather.

For the production of the electric light, Duboscq's lamp was used, set in motion by a battery of fifty small Grove's elements, and with this source of light photographs can be taken with any power with which pictures are possible in sunlight. The rubber cups of the battery were $4\frac{3}{4}$ inches high, $3\frac{1}{4}$ inches wide, and 2 inches thick; the platinums $5\frac{1}{2}$ by $2\frac{1}{8}$ inches, and weighed about 60 grains each. The zincs were bent on themselves so as to present a part of their surface on each side of the platinums, and weighed when new about a pound each.

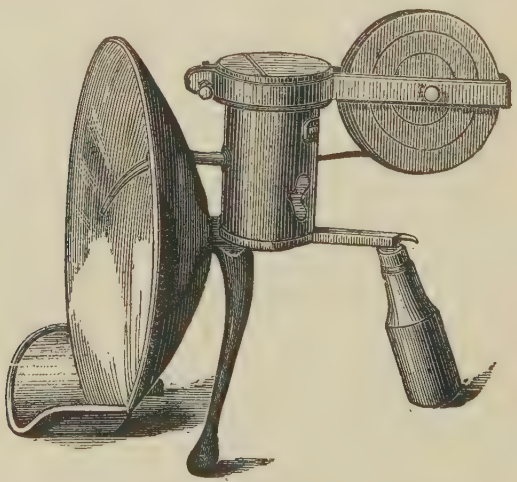


FIG. 198.

The electric light as hitherto employed has been a cumbrous, expensive, and troublesome process, which certainly could not be employed by the majority of persons wishful to practise photo-micrography. For those who may have to use high powers, and to whom daylight is not accessible, the magnesium light seems to possess

advantages. For this process a good lamp is necessary, and the one shown in Fig. 198 is the best of its kind.

In describing the magnesium modification of photography as applied to the amplification of microscopic objects, Colonel Woodward compares the light given off by a good magnesium lamp to white-cloud illumination of the best character.

Without the use of ground glass, this light serves admirably for the production of photographs of soft tissue with any power under 1000 diameters. The light being composed of a mixed pencil, with rays passing in all directions, there are no interference phenomena; but for the same reason, on the Nobert's plate and many test-objects, the results are inferior to those produced by the sun or by the electric light; with powers much higher than 1000 diameters, however, the time of exposure becomes inconveniently long.

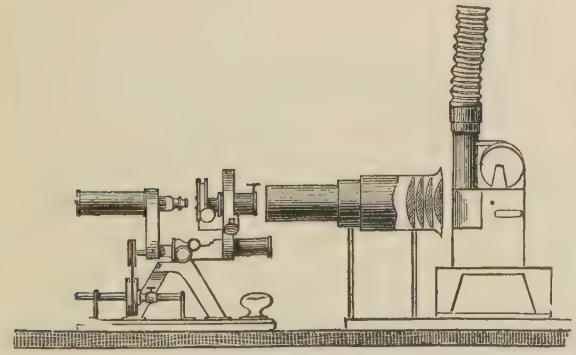


FIG. 199.

The condenser concentrates the light on the lower lens of the achromatic condenser of the microscope as shown in Fig. 199, which stands on a table supported by three levelling screws. The image is received on the plate-holder supported on the table, and photographed precisely

as when using the sun's rays as before described. The same focussing apparatus is employed, and the ammonio-sulphate cell should invariably be inserted, but the ground glass is never necessary. To produce negatives of tissue-preparations with 500 diameters it requires exposures of about three minutes. Other powers require proportionate exposures.

Magnesium wire costs 12*s.* 6*d.* per ounce; two ounces will, with care, answer for three or four hours' constant work, and ought to produce from twelve to thirty negatives, in accordance with the difficulties of the subjects to be represented. The fumes of magnesia resulting from the combustion are carried into a chimney five feet long, made of a spiral wire covered with muslin, terminating in a muslin bag, shown in the figure, in which the oxide condenses, while the draught goes on through the interstices of the muslin.

The method of photographing such objects as *Amphipleura pellucida* and Nobert's test-lines has also been described by Dr. Woodward in a letter to M. Deby, a vice-president of the Belgian Microscopical Society. Such objects as the *A. pellucida* require the use of very oblique light, which is obtained in the following manner: A parallel pencil of solar rays, from the heliostat and plane mirror, is intercepted by a blue cell and diaphragm, which only allows a circular pencil of half an inch diameter to pass. The light enters parallel to the optic axis of the microscope, placed in the usual position for photography, but at the lateral distance to the right or left of 3 inches. If the light is intercepted by a large achromatic prism, of a focal length of about 3 inches, the desired obliquity can be obtained without difficulty. The best result is obtained when the rays are concentrated to a focus upon the object, and it is indispensable that the stage of the microscope be as thin as possible. The illumination thus

produced is in general sufficient to produce negatives by the wet process up to 2500 diameters with three minutes' exposure. Fig. 112 shows that difficultly resolvable diatom the *A. pellucida*, under an amplification of 960 diameters.

After the sensitive plate has been exposed the necessary length of time by either of the foregoing methods of illumination, it is ready for development. It is, therefore, taken into a room from which every trace of actinic light has been excluded, and there the picture is brought to view by the use of the developing solution. Nearly every photographic manipulator has his own way of developing, though the general principles are the same, but that given by Messrs. Wratten and Wainwright for their instantaneous dry plates will perhaps be found the most suitable for photo-micrographical purposes.

STOCK SOLUTION A*.

Ammonia liquor, *fort.*	1 oz.
Potass bromide	60 grains
Water	2 oz.

DEVELOPER.

Pyrogallic acid	6 grains
Stock Solution A*	20 drops
Water	2 oz.

Lay the exposed plate in a dish of cold water to soak while the pyrogallic acid is mixed. For each $\frac{1}{4}$ or 5 × 4 plate use 6 grains of pyro., diluted with 2 oz. of water. First pour off the water from the plate and apply the pyro. solution, then add five drops of stock solution A*, and keep this weak developer on the plate until the highest lights are pretty well visible; then add from 15 to 20 drops more of A* to finish development. By this method more of the film is employed and greater density is obtained. Whenever any of solution A* is to be added to the pyro. solution it should be first dropped into the developing cup,

and then, if the solution which is in the dish be poured back to the cup a perfect admixture will be the result without the necessity of stirring. With correct exposure full printing density may be easily attained with the alkaline pyro. alone. No plate is supplied that will not bear 100 drops of stock solution A* without fog if the correct exposure has been given. One thickness of deep ruby glass is unsafe for the dark room ; two, at least, are necessary. Should any discoloration of the film appear after the negative has been fixed and washed, it may be cleared away with a weak solution of perchloride of iron and water (about the colour of pale sherry), the plate afterwards to be washed thoroughly.

Ferrous Oxalate Development.—Where it is not expressly stated to the contrary, Swan's plates may be developed either with ammoniacal pyrogallic acid or with ferrous oxalate ; but it occasionally happens that one and not the other of these developers must be used. When that is the case, the fact is stated on the label of the packet.

Ferrous oxalate developing solution is best made as follows :—

SOLUTION A.

Neutral oxalate of potash	6½ oz.
Water	25 oz.

SOLUTION B.

Sulphate of iron	2½ oz.
Water	7½ oz.
Sulphuric acid	1 to 2 drops.

The two solutions A and B are kept in stock, and when required for development three volumes of A are mixed with one volume of B.

It is better to mix and use what is immediately required for the development of a single plate, and not to use the same solution for developing several plates. The two stock

solutions keep well, therefore any quantity of these can be kept ready for mixing. The sulphuric acid is not essential in the formula referred to; it can be omitted.

The bromide of ammonium solution may be added to the developer as a restrainer, to counteract over-exposure in the same manner and under the same conditions as in development with pyrogallic acid and ammonia.

This ferrous oxalate developer should be used in a flat bath, precisely in the manner described for alkaline pyrogallic acid development. During the operation the solution must be made to wave to and fro over the plate, and the development continued until the required density is obtained. This will occupy about three minutes, as a rule. The addition of bromide affects most powerfully the rate of development and its character, making it slower and increasing the contrast of light and shade. For copying engravings, &c., where only black and white is to be printed, the proportion of bromide of ammonium may be advantageously increased.

If during development or fixing any loosening or blistering of the film should be observed, the plate must be immediately rinsed in water and put into a solution of alum (1 oz. to 1 pint of water). After allowing to remain five minutes in the alum solution, it should then be finished as usual after this treatment; and as all plates of the same packet will probably be alike, it will be advisable with each of these to use the alum bath after development, without waiting for the appearance of blisters or loosening of the film.

The image generally makes its appearance in about a minute, the deposit gradually increasing in intensity until the development is finished. It is then to be well washed and fixed in the following solution:—

Hyposulphate of soda	4 oz.
Water	20 oz.

In cases where, from over-exposure, it has been impossible to obtain density with the alkaline developer, a most careful washing should be given after fixing, with a view to remove the last trace of ammonia.

Intensification can then be effected with acid pyro. and nitrate of silver, or with protosulphate of iron and nitrate of silver. The author has a decided preference for the latter, and uses the following formula, viz. :—

A.

Protosulphate of iron	15 grains
Gelatino-acetic acid solution (as described below)	40 drops
Water	1 oz.

B.

Nitrate of silver	10 grains
Acetic acid, glacial, 50°	10 drops
Water	1 oz.

The gelatino-acetic acid solution is compounded as under :—

Gelatine	15 grains
Acetic acid, glacial, 50°	3 drachms
Water	5 drachms

and it is well to prepare a stock of this, and also of A, as they are both better for keeping.

To proceed—first flood the plate with water and then with a solution of iodine and iodide of potassium of the colour of *pale* sherry for one minute, rinse it off, and apply enough of A to cover the plate, for about the same time. Now drop into the cup a drachm of B, and bring the A back from the plate to the cup to mix them together. Re-apply, and keep moving over the surface until density is sufficient. If any air-bells should occur they must be kept moving, and then they will do no harm.

Both development and intensification are best performed in a dish—the former in an ebonite, the latter in one of porcelain.

When the plate has again become dry, warm and varnish it as usual.

Fixing.—It is well to finish fixing each plate by immersing it for about ten minutes in a perfectly fresh hyposulphite solution after the plate has, in the ordinary sense of the word, been fixed—that is, after the creamy appearance of the film has been discharged by immersion in a solution perhaps not quite fresh. The temperature of the hyposulphite solution should not be below 50° nor above 70° F. The apparent change in the strength of the image which attends fixing has led some to suppose that the image is greatly weakened by the action of the hyposulphite solution; but this is not the case, and we think it important to state clearly that the hyposulphite solution will not injure the image, even if it be allowed to act for a very much longer time than is necessary for the thorough fixation of the image and the removal of instable silver compounds from the film. Time must be allowed, both for the action of the hyposulphite of soda solution, and also for thorough washing afterwards, in order to effect the complete removal from the plate of compounds which, if not removed, will ultimately destroy the plate.

Finishing the Plate.—The plate must be dried spontaneously. On no account may artificial heat be applied. In case of urgency drying may be hastened by immersion in methylated spirit, and then varnished when dry. A suitable varnish for this purpose is Mawson and Swan's "dry plate varnish." In every case the negative should be carefully preserved from damp. Enamel collodion is a useful temporary varnish for negatives on gelatine plates.

Having obtained a suitable negative, it is easy to print from this either upon carbon tissue or the ordinary sensitised paper, which latter may now be bought ready prepared at 1*s.* per sheet. It will keep in good condition for months.

After printing several shades darker than is finally required, the prints are soaked in water to remove the free nitrate of silver, and then put to tone in one of the following baths :—

I.

Chloride of gold	15 grains
Acetate of soda	1 oz.
Water	80 oz.

II.

Dissolve one 15-grain tube of chloride of gold in 2 oz. of water. Take 1 drachm of this solution, add 5 grains of tungstate of soda and pour in 3 oz. of boiling water. Use the bath ten or fifteen minutes after mixing ; one grain of gold chloride will tone a sheet of paper.

III.

Chloride of gold	15 grains
Chloride of lime	15 grains
Precipitated chalk	1 teaspoonful
Water (boiling)	80 oz.

This may be used at once.

IV.

Chloride of gold	7½ grains
Perchloride of platinum	7½ grains
Acetate of soda	1 oz.
Water	80 oz.

After toning, the prints must be well washed and transferred to the fixing solution, which should be of the same strength as that used for fixing the negatives. The prints should be allowed to remain in this solution for ten minutes, after which they are transferred to water, and washed until perfectly freed from all traces of the hyposulphite.

There are two applications of this art which may be of great utility, viz. the preparation of wood-blocks for the purpose of illustrating microscopical work and also for the production of lantern transparencies. The greatest expense

in producing good illustrations is that of the artist, but if the picture is put upon the wood the remainder of the work is cheaply and expeditiously performed. One way of doing this is to transfer an autotype carbon positive to the prepared wood-block; but to produce good pictures upon wood it is better to employ the wet collodion process.

A slightly over-developed positive is produced upon glass in the ordinary way, being developed with an iron developer, and the film transferred to the wood-block in the following manner: The block is first to be coated with a gelatine solution made by soaking 1 oz. of Nelson's gelatine in 19 oz. of water for twelve hours, dissolving with heat and stirring in a solution of 20 grains of chrome alum in 1 oz. of water. When thoroughly incorporated, add sufficient lampblack to form an even black coating upon the wood.

If the block be first coated, it will be dry and ready to receive the film by the time the operator has proceeded thus far. The positive having been taken, developed, and fixed, a piece of gummed paper is "squeezed" to the wet film, which must then be stripped off the glass. The paper and film may now be trimmed with the scissors, and brought into contact under water with the prepared block, gently squeezed to remove air and water, and set aside for a while, so that the two films may become amalgamated. After a short time the block is soaked in tepid water to remove the gummed paper, and after drying is ready for the engraver. Many of the illustrations in this work have been prepared by this process, and it is here recommended to all who may read papers before the members of the various societies with which they may be connected.

In the preparation of transparencies for the lantern, it is necessary first of all to secure a good and sharp nega-

tive. This is placed above a dry plate, the two films being in contact, exposed to the light in a printing frame, and the latent image developed and fixed in the usual way. The best dry plates with which to prepare transparencies are the collodio-albumen, developed with pyrogallic and citric acids, and next to these, any good collodion plate. It is a very difficult matter to prepare *brilliant* transparencies upon gelatine plates; they are so extremely sensitive to light that it is next to impossible to keep the high lights clear and free from fog. Very passable pictures may, however, be obtained from them by giving 12 seconds' exposure, under a negative of ordinary density, to the light of a common bat's-wing gas-burner.

The collodio-albumen plates, when developed with pyrogallic and citric acids, generally yield a fine blue-black image; they may, however, be readily toned in a *weak* solution of sulphide of ammonium, if requisite.

When dry the picture should be covered with another piece of clear glass the same size, the two being prevented from touching by very small strips of card fastened by gum to two of the edges, the whole bound round with thin black paper, and the slide is completed. These pictures, when laid upon white paper, should only allow the paper to show through in the very highest lights, just a patch of white here and there.

For the illustration of papers read before societies, transparencies prepared by the foregoing method may be shown by the use of the Sciopticon or Triplexicon (Fig. 195), by which means sufficient illumination can be obtained with paraffin oil to produce a good disc 6 or 7 feet in diameter. It is, of course, necessary by this method to first produce an enlarged transparency of the object, which entails some previous trouble, though it is less costly than that next to be described.

Mr. Browning has prepared a nose-piece for lanterns, which he calls the "Lantern Microscope," by the use of which the ordinary 3 × 1 microscopic slides may be displayed upon the screen. Its form may be seen in Fig. 200.

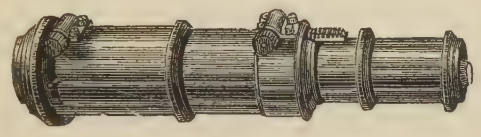


FIG. 200.

It is composed of the following parts, conveniently mounted in brass, with rack adjustments and other requisites, and so arranged that it can immediately replace the ordinary front combination of first-class lanterns:—(1) Lenses acting as an achromatic condenser, receiving the light that passes through the large lenses of the lantern and reducing the light-pencil to the proper size. (2) An object-holder for 3 × 1 inch slides. Then follows the magnifying apparatus, as in a table microscope—namely, an achromatic objective, made specially for this instrument, and a Kelner eye-piece, giving considerable amplification with a very wide field. The light-pencil must be adjusted by means of the rackwork attached to the second condenser, and an image of the object to be displayed must be received on a well-strained *paper* screen, and carefully focussed by turning the milled head furthest from the object. The objects best suited for display are transparent injections, which should be thin and not too darkly stained; entire insects, mouth organs, tracheal tubes, &c., of the larger kinds, sections of wood, tissues and organs of plants, preparations of zoophytes, sections of coal and other minerals. To prevent disappointment, it should be remembered that transparent objects, such as diatoms and the

smaller infusoria, do not give sufficiently positive outlines for this method of display. Neither oil lamps nor the paraffin lamp of the Sciopticon or Triplexicon yield sufficient light. For magnifications of from 50 to 100 diameters lime-light will perform well, but for higher powers the electric light should be used.

MICROMETRY.—The micrometer, as its name signifies, is used for measuring small objects, and omitting all historical notices—which may, however, be found in the works of Baker and Quekett—we will proceed to describe those forms most in use.

FIG. 201.

The stage micrometer consists of a glass slip such as is shown in Fig. 201, upon which are ruled a series of lines generally $\frac{1}{100}$ of an inch apart, one of these divisions being again divided into ten parts; so that there is on the same scale the means whereby objects of varying magnitude may be exactly measured, to less even than the thousandth of an inch.

Some micrometers are also ruled to *millimetres*, tenths and hundredths, and this system is daily coming more into use. A standard system for measurements was long required for scientific microscopical purposes, and by very general consent the thousandth part of a millimetre has been taken as this standard, and by some termed a *micro-millimetre;* while others, objecting to the prefix "micro," which in electrical measurements means a millionth of the unit, prefer to call the unit of the thousandth of a millimetre a *micra*. One thousand micras make one millimetre. The following table, extracted from the 'Journal of the

Royal Microscopical Society,' may be of use to the reader:—

Fractions of an inch.	Micras.	Fractions of an inch.	Milli-metres.	Micras.	Decimals of an inch.
	μ		mm.	μ	in.
1/10000	2·539977	1/900	·028222	1	·000039
1/9000	2·822197	1/800	·031750	2	·000079
1/8000	3·174971	1/700	·036285	3	·000118
1/7000	3·628539	1/600	·042333	4	·000157
1/6000	4·233295	1/500	·050800	5	·000197
1/5000	5·079954	1/450	·056444	6	·000236
1/4000	6·349943	1/400	·063499	7	·000276
1/3000	8·466591	1/350	·072571	8	·000315
1/2000	12·699886	1/300	·084666	9	·000354
1/1000	25·399772	1/250	·101599	10	·000394
		1/200	·126999		
		1/150	·169332		
		1/100	·253998		

The stage micrometer cannot be used for *direct* measurements with any save low powers, as it is necessary that the lines be seen at the same time as the dimensions of the object; but indirectly very exact measurements may be obtained. This may be accomplished by sketching the outlines of the object lying upon the stage, using the camera-lucida as shown in Fig. 189. The object is then to be replaced by the stage micrometer, the lines of which are drawn over the previous sketch, showing by simple inspection the magnitude of the object, or any part of it may now be measured with a pair of compasses.

It should be borne in mind that amplifications are generally expressed at a distance of 10 inches, and this is accomplished by placing the drawing slab horizontally 10 inches beneath the acute angle of the camera-lucida prism. If the object is required to be of the same size as it appears upon looking into the microscope, the distance of the drawing slab from the acute angle of the prism must be equal to the distance of that point from the objective. In estimating the magnifying power of an objective, the stage

micrometer is employed and a sketch made of it upon the drawing slab at a distance of 10 inches. If, upon measuring these lines with an ordinary rule divided into inches and tenths, the hundredth of an inch space corresponds to one linear inch, it is clear that the object has been magnified 100 diameters; while if the thousandth of an inch space occupied half an inch on the rule, it would have been amplified 500 diameters.

Eye-piece micrometers are of several kinds, the simplest being a circle of glass ruled with a series of squares $\frac{1}{25}$ to $\frac{1}{100}$ of an inch (or $\frac{1}{10}$ of a millimetre) apart, being made to rest on the diaphragm of the eye-piece, or placed in front of the field lens in Ramsden's pattern. Previous to using this form of micrometer the value of the sides of these squares, with each objective, will have to be determined; this is done in the following manner:—Place a stage-micrometer, ruled to hundredths and thousandths of an inch (or tenths and hundredths of a millimetre), in position upon the stage, and view it with the eye-piece having the ruled circle *in situ*. The lines upon both micrometers must now be rendered parallel, and carefully observed where the one overlaps the other, and if any uneven divisions appear, it will be better to pull out the draw-tube until they become even, keeping a record as to how much the tube has been withdrawn. As an instance—when using the 1-inch objective and an A eye-piece, each $\frac{1}{100}$ of an inch of the stage micrometer *nearly* coincided with ten divisions of the ruled glass circle. In order to make the coincidence exact, the draw-tube was pulled out $\frac{3}{10}$ of an inch; therefore, for that objective and ocular, each division of the eye-piece micrometer represents $\frac{1}{1000}$ of an inch, when the draw-tube is extended the same amount.

Jackson's eye-piece micrometer is shown in Figs. 202

and 203, inserted into the ordinary Huyghenian eye-piece. The micrometer itself consists of a series of lines ruled upon glass and set in a frame so that it may be moved forward by a small screw, a spring at the extreme end keeping it always in contact with it. The slits which are cut in each side of the eye-piece should be provided with a small sliding tube to prevent dust entering between the glasses.

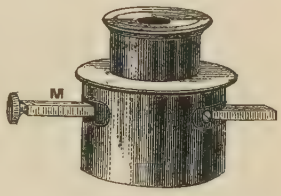


FIG. 202.

The method of finding the permanent values of the divisions of this micrometer is identical with that previously described, and when once found with accuracy should be entered in the observer's note-book for future reference.

FIG. 203.

Ramsden's screw micrometer, the last we need mention, is shown in Fig. 204. It is composed of a positive eye-piece (see page 42), before which is stretched two very fine parallel wires, one of which can be separated from the other by a screw having one hundred threads to the inch, the head being further graduated into one hundred parts, as shown in the figure representing the instrument made by Mr. Swift.

The field of view is illustrated by Fig. 205, and shows clearly the lower portion, consisting of a thin plate of brass, the edge being indented with a series of notches; every fifth indentation is deeper than the others, thus facilitating the counting. Each notch is equivalent to one

turn of the milled head, so that the number of revolutions may be read off without taking the eye from the object.

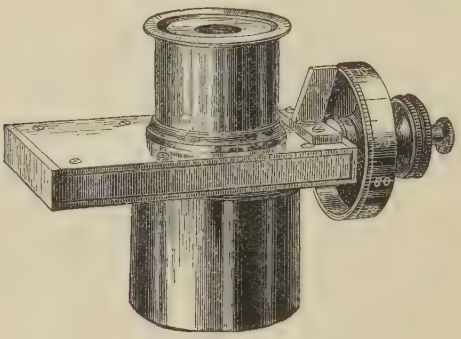


FIG. 204.

Quekett, in his 'Practical Treatise on the Use of the Microscope,' has shown the *theoretical* possibility of measuring a space so small as the one eight hundred thousandth of an inch with the eighth objective and this micrometer; but there are many practical difficulties in the way of attaining to this.

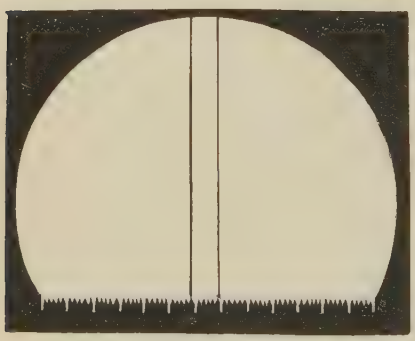


FIG. 205.

MEASUREMENT OF ANGULAR APERTURE. — Under the head of microscopic measurements may be described the method of using Prof. Abbe's apertometer for measuring the aperture of objectives, a rough method of accomplishing this having been already described on page 56. An illustration of this instrument may be seen in Fig. 206. It is a semicircular disc of crown glass, 3·5 inches in diameter, and half an inch in thickness, polished on the cylindrical edge and ground to an angle of 45° upon the

diametrical edge. The centre of the semicircle is formed by a small hole in a silvered cover *a*, cemented to the upper face of the disc. Two indices of blackened brass *b*, *b*, sliding on the periphery of the disc, afford visible marks for observing the aperture of any objective. On the upper face of the disc there are two engraved scales, the outer indicating the numerical aperture by direct observation.

FIG. 206.

In order to measure the aperture of any objective the apertometer is placed upon the stage of a microscope, and the objective, the aperture of which is to be measured, roughly focussed to the small hole *a*, in the silvered cover. In the case of an immersion lens, a drop of immersion fluid must of course be applied. This done, the eye-piece of the microscope is removed and the eye placed to the open tube, the two indices of blackened brass *b*, *b*, being moved to and fro until the points just touch the margins of the illuminated field. The position of the indices is then read off on the outer circle of figures, and half the sum of both these readings will give the aperture of the objective.

For lenses of higher power than the $\frac{1}{2}$-inch, a 2-inch objective is provided. This is made to screw into the lower end of the draw-tube and used with an ordinary A

eye-piece, the whole being moved up and down until the edge of the disc is seen quite distinctly; the procedure is then the same as with low powers. This auxiliary objective must be furnished with a diaphragm to cut off all false rays, the aperture being one-eighth of an inch in diameter, and situated 1·5 inches from the achromatic combination.

The numerical aperture divided by the refractive index of the medium in which it is measured produces the sine of the semi-angle, and thus the angle in any medium may be easily obtained.

The following table of natural sines may be of use in this direction:—

NATURAL SINES.

Degrees.	Sines.	Degrees.	Sines.	Degrees.	Sines.
1	·0175	31	·5150	61	·8746
2	·0349	32	·5299	62	·8829
3	·0524	33	·5446	63	·8910
4	·0698	34	·5592	64	·8988
5	·0872	35	·5736	65	·9063
6	·1045	36	·5878	66	·9135
7	·1219	37	·6018	67	·9205
8	·1392	38	·6157	68	·9272
9	·1564	39	·6293	69	·9336
10	·1736	40	·6428	70	·9397
11	·1908	41	·6561	71	·9455
12	·2079	42	·6691	72	·9511
13	·2249	43	·6820	73	·9563
14	·2419	44	·6947	74	·9613
15	·2588	45	·7071	75	·9659
16	·2756	46	·7193	76	·9703
17	·2924	47	·7314	77	·9744
18	·3090	48	·7431	78	·9781
19	·3256	49	·7547	79	·9816
20	·3420	50	·7660	80	·9848
21	·3584	51	·7771	81	·9877
22	·3746	52	·7880	82	·9903
23	·3907	53	·7986	83	·9925
24	·4067	54	·8090	84	·9945
25	·4226	55	·8191	85	·9962
26	·4384	56	·8290	86	·9976
27	·4540	57	·8387	87	·9986
28	·4695	58	·8480	88	·9994
29	·4848	59	·8572	89	·9998
30	·5000	60	·8660	90	1·0000

Abbe's apertometer is one of the instruments which may be profitably purchased by the executive of every microscopical society, for use in testing the apertures of the members' objectives. Such a proceeding would often save the individual member much annoyance, and he would then be certain of what he was purchasing. A case of this kind has just fallen under the author's notice. A $\frac{1}{8}$ objective was obtained of professedly 140° air angle, and 8*l.* 10*s.* paid for it, while, when it came to be accurately measured, it turned out only 108°, and such a one could have been bought for 50*s.* It should be the duty of the executive of every microscopical society to secure its members against fraud, and the method above stated would most assuredly do this.

CHAPTER X.

THE POLARISCOPE.

WHEN a ray of light is made to pass in a certain direction through a crystal of Iceland spar, it becomes split up into two portions of equal intensity, an *ordinary* ray and an *extraordinary* ray respectively. If a ray, having undergone ordinary refraction through a crystal of Iceland spar, be allowed to pass through a second crystal, it is split up in the same manner; but the ordinary and extraordinary rays will be of different degrees of intensity. When the second crystal is rotated until the two are in similar or opposite positions, then the ordinary ray appears at its greatest intensity, while the extraordinary ray disappears. If the second crystal is further rotated the extraordinary ray reappears and increases in intensity as the rotation proceeds, while the ordinary ray diminishes in intensity until the principal planes of the crystals are at right angles to each other, when the ordinary ray vanishes and the extraordinary ray appears at its greatest intensity.

These and similar phenomena are called polarisation, and two prisms are required to render it patent—the first for polarising the light, called the "polariser," or the "polarising prism;" and the second, necessary for exhibiting the fact that the light has undergone modification, called the "analysing prism," or simply the "analyser."

There are several methods of producing the polarisation of light, but the one usually employed for microscopical purposes is by the aid of the Nicol or single image prism, which yields a beam perfectly colourless, polarising com-

pletely, and transmitting but one of the rays, the other being entirely suppressed.

The Nicol prism is made from a rhombohedron of Iceland spar, bisected in the plane which passes through its obtuse angles, as shown in Fig. 207 by the letters A B C D, the halves being joined again in the same order by means of Canada balsam.

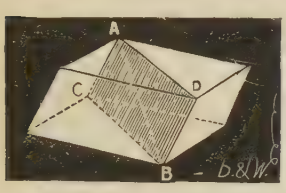


FIG. 207.

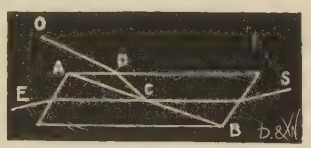


FIG. 208.

When a luminous ray enters this prism the ordinary ray undergoes total reflection, passing out of the crystal at O, while the extraordinary ray emerges at E, Fig. 208.

As attached to ordinary microscopes both polariser and analyser are formed of Nicol prisms; the former being made to fix underneath the stage or into the sub-stage. For this purpose it is mounted as shown in C (Fig. 209), so that the prism may be easily rotated by means of the milled head.

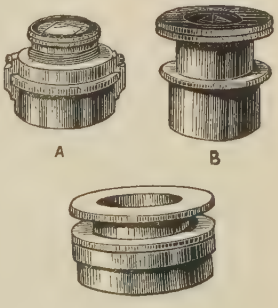


FIG. 209.

The analyser may be used in several ways. In the first, the Nicol prism is set in a mount screwing into the lower end of the microscope body, just above the objective, or in a brass box sliding into the side of the tube, in a similar manner to the Wenham prism of a binocular instrument. In another, the prism fits over the eye-piece in the place of the ordinary

cap, each kind of apparatus being shown in Fig. 209, where the eye-piece analyser is shown at B, the other form at A.

Sometimes a simple rhomb of Iceland spar, called also a "double image prism," is used as an analyser, but is not so generally useful as the Nicol prism previously described, though a series of interesting experiments may be made with it.

There are several other methods of polarising light: By means of the bundle of glass polariser, the plate of black glass, the tourmaline plate, and the sulphate of iodo-quinine crystal; but as all these can be seen in almost any treatise on optics, it seems scarcely necessary to more than mention them here.

The tourmaline plate may, perhaps, receive notice; in this crystal the optical axis coincides with the axis of the prism, and for optical purposes a plate is cut from it in a plane parallel to this axis. There are many objections to the use of a tourmaline plate, the principal being that a considerable thickness is required, as unless the ordinary ray is completely absorbed, the emergent light will be only partially polarised, and this requisite thickness produces a very undesirable colour.

When the analysing prism is placed immediately above the objective it stops a considerable amount of light, though it gives a full field, while when placed over the eye-piece a smaller field but brighter image is obtained.

It is often useful to produce certain colour effects when working with polarised light, and with this end in view a plate of selenite is interposed between the polariser and the object. Films of varying thicknesses are used; they are mounted in various ways, as selenite stages, object-carriers, and the like. Perhaps the best form of selenite stage is that made by Mr. Swift; it is shown in Fig. 210;

the reader will be able to make out its construction from the illustration.

Mr. Swift's complete condenser, which has been already shown at Fig. 44, is also fitted with selenite discs for colour

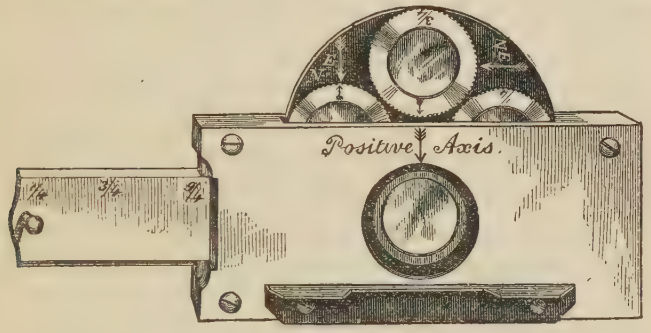


FIG. 210.

development, and also with a revolving film of mica, by the combinations of which a very great number of tints may be obtained.

Darker's selenite discs, as made by Messrs. Beck, are shown in Fig. 211, and are very useful, seeing that one or all of them can be put in and out of use in a moment.

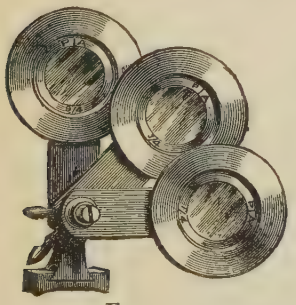


FIG. 211.

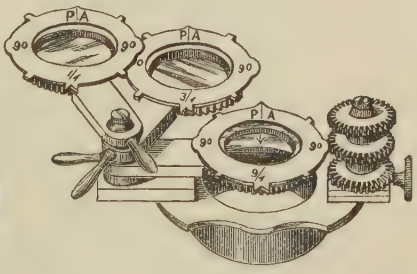


FIG. 212.

This form has been modified by Mr. Richards, who read a paper upon "A New Method of using Darker's Films" before the Royal Microscopical Society, March 9th, 1870.

His improved apparatus may be seen in Fig. 212, having been reduced from the drawing which appeared in the 'Monthly Microscopical Journal.'

The real advantage of the apparatus (if any) seems to be in the application of gearing to produce revolution of the films, an operation easily performed by the fingers alone in the Ross instrument, as well as those of other makers.

The colours produced by the selenite films are brought about by variations in their thickness, the red being yielded by the thickest, violet by the thinnest; these films vary between ·00124 and ·01818 of an inch. The arrow-head and letters P A indicate the positive axis of the selenite, while the figures $\frac{1}{4}$, $\frac{3}{4}$, and $\frac{9}{4}$ denote the parts of a vibration retarded by each disc.

But the student need not go to much expense over these elaborate stages. They are intended to save time and not money, and the ordinary plain mounted selenite films on a 3 × 1 inch glass slide will answer many purposes, though incapable of rotation. If the red and green, blue and yellow slides be provided, with a film of mica, this is about all the student need purchase, at all events for some time to come.

Rock crystal or quartz possesses the extraordinary power called rotatory polarisation. If a ray of homogeneous light be polarised, and the analyser turned so that it appears totally extinguished, the ray can be made to pass again through the analyser by placing a thin section of quartz, with its plane at right angles to the rays, between the polariser and analyser. In order to effect this the quartz crystal must be cut at right angles to its axis.

In most petrological microscopes of the present day Klein's plate of quartz is inserted in a movable box placed

immediately above the objective. This plate is usually 3·75 millimetres in thickness, and is of the greatest utility when very feeble doubly refracting crystals are being examined, and in many other instances where it is suspected that isotropic particles exist in admixture with amorphous matter of a doubtful character.

The action of two Nicol prisms with an interposed plate of quartz upon a beam of light has been made use of for the purpose of registering the position of absorption bands in the spectra of various substances; but this properly belongs to the next chapter.

The development of the study of objects by the aid of polarised light may in the end bring about important discoveries, and therefore should be studied more carefully than it is at present.

Let the student examine some potato starch by the use of ordinary light, and after noticing the position of the hilum, again examine with polarised light, when he will not fail to notice the black cross, as shown in Fig. 213.

FIG. 213.

For the observation of starches under polarised light, they should be mounted in gum dammar dissolved in benzol—as in aqueous fluids the effect is not developed in so marked a manner. Wheat starch, which only faintly shows the cross when observed in water, exhibits it very plainly in gum dammar and benzol. The effects of polarised

light on the various starches may be seen in Fig. 145. The palates of molluscs are displayed remarkably well under the influence of polarised light, especially when mounted in gum dammar or Canada balsam.

A very common object, seen in almost all collections of slides, is the section of rhinoceros horn. When examined by ordinary light it appears to resemble a bundle of hairs; but when viewed with the polariscope, each cylindrical aggregation is marked by a cross, bearing a striking resemblance to the starch granules illustrated by Fig. 213.

The muscular system of many minute animals can only be satisfactorily made out by means of polarised light, which very strikingly differentiates the tissues; while such objects as silk, cotton, flax, and jute are often to be distinguished from other substances by its aid.

But by far the most important application of the microscope and polariscope is in relation to petrology, whereby much regarding the nature of most minerals may be satisfactorily made out. With this end in view several petrological microscopes have been produced during the past few years, notably from the houses of Mr. Watson and Messrs. Swift and Son, the latter making the instrument shown in Fig. 214.

This instrument is specially designed for the examination of rocks and mineral substances, being furnished with a concentric and revolving glass stage, self-centering arrangement, goniometer, and complete polariscope. The polarising prism is also provided with a divided circle and spring catch to indicate when the Nicols are crossed; it is made to slide up in its fitting close to the under side of the object, and on it are fitted condensing lenses of extreme high angle for showing the brushes in crystals. When not required, the polariser can be moved aside, entirely clear of the stage of the microscope. The analyser is made to slide in the

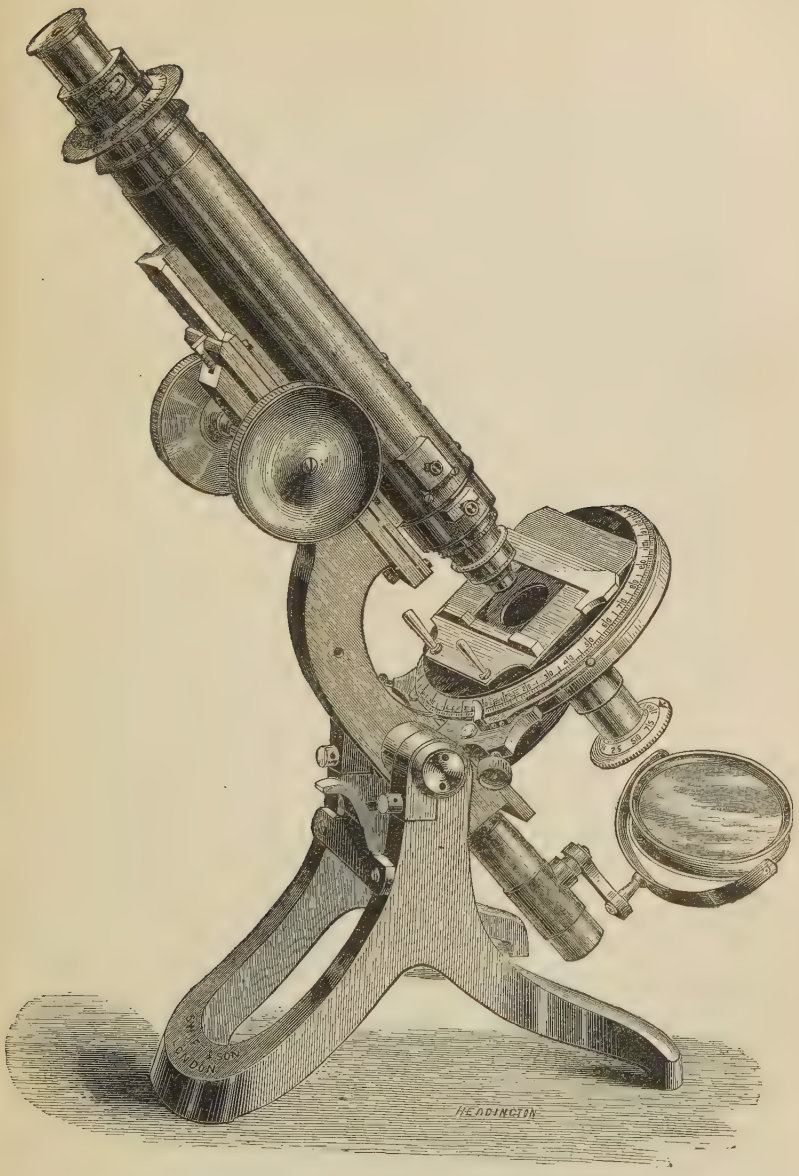


FIG. 214.

optical tube, immediately under which, in the same way, is fitted a Klein's quartz plate. An extra analysing prism with divided circle is placed over the eye-piece, and a contrivance for rotating crystals between it and the prism is also added. The eye-piece has spider lines accurately adjusted to the plane of the prism.

When the Nicol prisms (polariser and analyser) are set at right angles to each other they are said to be crossed, and there is a total extinction of light passing to the eye of the observer; a thin section of an amorphous substance, such as glass, if placed upon the stage, does not produce any change, neither does a similar slice of any mineral which crystallises in the cubic system; and if the field remains perfectly dark when an object is observed between crossed "Nicols," it may be safely inferred that it belongs to one of these classes, or happens to be lying in a plane yielding single refraction only.

Some crystals appear not to polarise until the stage upon which the slide is laid be rotated, and if it becomes visible by this treatment it is certain the crystal does not belong to the cubic system.

Minerals of all the other systems of crystallography behave in various manners under crossed Nicols, and this branch of study may be easily carried out with the aid of the information to be found in Chapter VIII. Sections should be cut in various planes of the crystal so as to represent the principal directions of vibration and also the axes of elasticity; but as this properly belongs to the science of petrology, the student should refer to those treatises in which these questions are specially considered.

In the petrological microscope shown by Fig. 214 there is fitted an arrangement for showing the rings in biaxial crystals: a crystal of augite spar (*diopside*) is brought into the field of the microscope, and its entire system of rings

may be exhibited as large, and with as much brilliancy, as in Norremberg's polariscope.

These coloured rings form a curious study; when, for instance, a section of Iceland spar (a negative uniaxial crystal) is cut from 1 to 20 millimetres thick in a plane perpendicular to the axis, and observed with polarised light, the following curious appearances are presented. First, when the axes of both polariser and analyser are perpendicular, there is shown a beautiful series of coloured rings traversed by a black cross; and secondly, when the axes are parallel, the coloured rings possess complementary tints to those shown at first, while a white cross has taken the place of the black one.

In biaxial crystals the system of rings is more complicated, and this series may be well studied in a plate of nitrate of potash, cut perpendicularly to its axis.

In order to exhibit the phenomenon of double refraction a "double-image prism" is used over the eye-piece, in conjunction with an objective; a plate of brass, pierced with a series of holes $\frac{1}{16}$ to $\frac{1}{4}$ of an inch in diameter, being laid upon the stage. When the polariser is fitted into its place below the stage, and the double-image prism over the eye-piece, there will appear (in some positions) two images, while when the prism is moved, one of the images will be completely obliterated, a change taking place at every quarter of a revolution. When the polariser is taken away the eye-piece shows two distinct images of the light circle, the ordinary and the extraordinary ray, which describe a circle upon revolving the eye-piece. If one of the larger apertures be now placed under the objective the images will not be completely separated but will overlap.

These are only a few of the many curious effects produced by polarised light, and its *study* should be more fostered and encouraged.

It is not enough to mount slides of salicine and other crystalline forms for the production of rainbow colours, and therefore the author imagines he cannot do better than give a list of the principal works which may be consulted:—'Lectures on Polarised Light,' by T. P., Longman and Co., 1843; 'Polarisation of Light,' by William Spottiswoode, Nature Series, 1874; 'A Familiar Introduction to the Study of Polarised Light,' by C. Woodward, Van Voorst, 1861; 'Minéralogie Micrographique,' MM. Fouqué and Lévy, published by the French Minister of Public Works, and is a most valuable work to all concerned in microscopic mineralogy.

To conclude this chapter a list is given of many polarising objects:—

ANIMAL SUBSTANCES.

Palate of limpet.	Human corns.
,, periwinkle.	Horn of rhinoceros.
,, whelk.	,, antelope.
Embryo oysters.	Skin of rhinoceros.
Human hair.	Alpaca wool.
Raw silk.	Porcupine quill.

VEGETABLE SUBSTANCES.

Starch from tous les mois.	Plant raphides.
,, potato.	Seed of Eccremocarpus.
,, arrowroot.	Cuticle of *Correa cardinalis*.
,, maize.	Fern scales.
Cotton fibres.	Jute fibres.

CHEMICAL CRYSTALS.

Salicine.	Asparagine.
Quinine.	Quinidine.
Chlorate of potash.	Strychnine.
Cane sugar.	Tartaric acid.
Brownian motion in fluids.	Citric acid.
Oxalic acid.	Sulphate of lime.
Sulphate of copper.	Chloride of barium.

CHAPTER XI.

THE MICRO-SPECTROSCOPE.

THE micro-spectroscope is used as a means for exhibiting the absence of certain rays of light in a beam which has been modified by its passage through certain substances or solutions. Generally speaking there are two patterns of the instrument, the former made by Messrs. Beck, as shown in Fig. 215, and that made by Mr. Browning, shown in Fig. 216. There is not much difference between the two in actual working, the form the author prefers, however, being that of Mr. Browning.

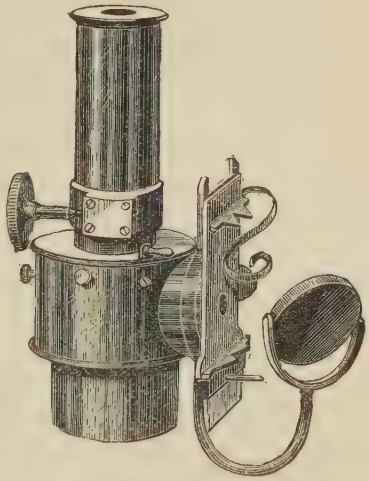


FIG. 215.

The Sorby-Browning instrument consists of a series of prisms, of crown glass and flint, mounted in a small tube, so that they can be removed at pleasure. Immediately below this tube is an achromatic eye-glass; and below the adjustable slit, placed about the centre of the widened part of the tube, is an ordinary plano-convex field-lens.

The amount of dispersion, that is, the length of the visible spectrum, is produced by alterations in the prisms, and it is as well for the worker in this branch of micro-

scopy to be provided with several copies of the tube, containing prisms yielding different degrees of dispersion. Mr. Browning has made for the author four such tubes, producing a small, medium, and large amount of dispersion, one of which can easily be substituted for the other.

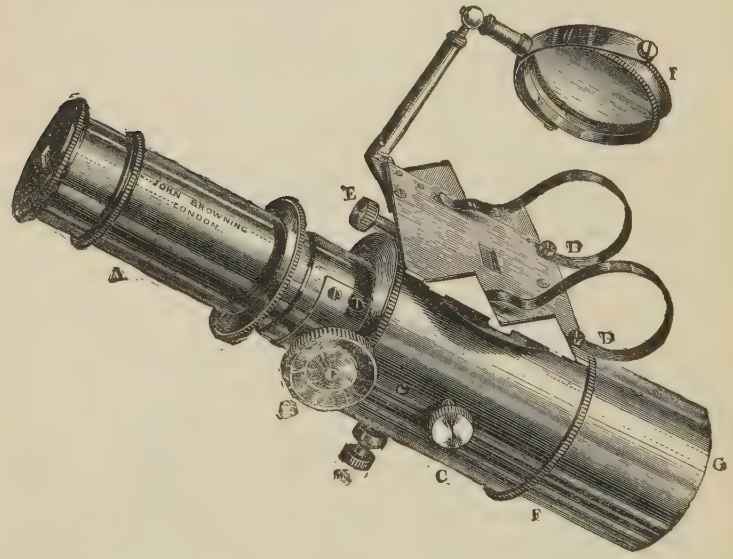


FIG. 216.

The student must remember that an increased dispersion does not define the absorption bands any the better; on the contrary, fine and delicate lines, easily seen by the use of a prism with moderate dispersion, are often lost when one of large dispersion is substituted.

To use this instrument, insert it as an eye-piece in the microscope, screw on the object-glass, and place the object, the spectrum of which is required, upon the stage. Illuminate by means of the sub-stage mirror if the object is transparent, or with mirror, Lieberkuhn, or side-reflector if opaque. Remove the tube A, and open the slit by means

of the milled head H. Focus the object and then gradually close the slit till a good spectrum is obtained, and this may be further improved by throwing the object a little out of focus. Each portion of the spectrum differs a little from adjacent parts in refrangibility, and delicate bands or lines can only be brought out by focussing their own parts of the spectrum, which may be done by means of the milled head B.

One advantage which the micro-spectroscope possesses is that extremely small quantities may serve for observation, and for this purpose powers varying from $\frac{1}{2}$-inch to $\frac{1}{20}$-inch may be used.

It is very easy to have the tints of the solutions examined too intense, so that nothing but a general and indefinite absorption appears throughout the entire spectrum. Nothing, perhaps, can show this better than a solution of permanganate of potash or Condy's pink fluid.

Mr. Sorby has devised what he has termed a wedge cell, in which to view the liquid for its absorption; it is shown in Fig. 217. By this means an increasing thickness of fluid and consequent intensity of colour can be brought under observation with the minimum of trouble. Again, very

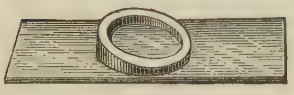


FIG. 217.

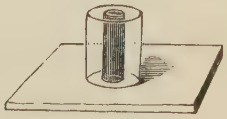


FIG. 218.

minute quantities of solutions may be examined in the slide shown in Fig. 218, made of a piece of barometer tube cut and ground evenly, and then cemented to a piece of ordinary glass.

Mineral sections, crystals, blowpipe-beads, &c., should

invariably have a small cardboard diaphragm, one-eighth diameter, placed beneath them; the spectrum is then much better defined. With a slide containing a mass of small crystals the object need merely be thrown a little out of focus.

When observing the spectra of liquids in experiment cells, or through small test-tubes, always slip over the 1½ or 2-inch objective a cap with a hole $\frac{1}{16}$ of an inch diameter. Slide this cap just sufficiently to bring the small hole a little within the focus of the objective. By this arrangement all extraneous light is prevented from passing up the body of the microscope, except what passes through the object. Unless this precaution be attended to, a false result is sometimes obtained.

Substances which give bands or lines in the red are best seen by gaslight, while those which give bands in the blue are brought out far better by daylight. Such a specimen as oxalate of chromium and soda is almost opaque by daylight, showing no bands, though when examined by a lamp the spectrum exhibits three beautifully fine lines in the red, two of which are exceedingly delicate. Again, uranic acetate can only be seen to advantage by strong daylight, since the band in the violet would be invisible by lamplight.

In solids, as crystallised uranic acetate, the tubes should be laid over a piece of black velvet on the stage of the microscope and illuminated with a bull's-eye condenser or side reflector.

It may be at first thought that absorption in the spectrum is due to the particular colour only—that is to say, a blue would always act as the ammoniacal sulphate of copper; a brown, as Parisite; a green as chlorophyll; and all reds would absorb the same light from the spectrum as a film of blood; but a few experiments would soon show

that this is by no means the case, and a reference to Fig. 219 will show this clearly.

In the illustration, A represents the absorption spectrum of oxalate of didymium viewed by reflected light. The oxalate is a white powder, yet the six bands are very plainly defined even with ordinary instruments, while with a good set of prisms the broad band at the red end of the spectrum may be resolved into several dark lines.

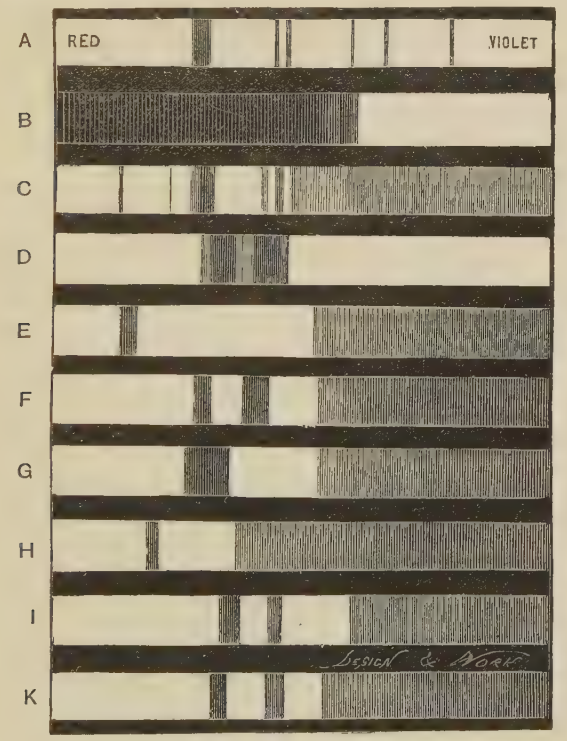


FIG. 219.

The absorption bands of didymium were first described in 1858 by Dr. J. H. Gladstone, and are remarkable for their extreme sharpness.

At B is shown the spectrum of the deep blue solution

of ammonio-sulphate of copper, already alluded to as being used for purposes of photomicrography, and the figure here shows us how completely the red end of the spectrum is cut off, and how little of the actinic portion is absorbed. C is given to show how mixtures of substances may be detected; the illustration represents the absorption bands of Parisite—a brown and somewhat rare mineral from the emerald mines of New Granada. It consists principally of the carbonates of cerium, lanthanium, and didymium, the spectrum of the last showing out with its usual distinctness. At D may be seen the spectrum produced by a colouring matter obtained from the wing feathers of the Cape lory (*Turacus albocristatus*) by Professor Church, and to which he has given the name "turacine." E shows the spectrum (or one of them) of chlorophyll, and is characterised by the deep broad band in the red end of the spectrum; K, the orange colouring matter obtained from aphides by Mr. Sorby, remarkable on account of the band in the yellow. F, G, H, and I form a series; they are the spectra of blood and its modifications. F is fresh blood; G deoxidised blood; H blood treated with citric acid; while I is similar to H but deoxidised.

Mr. Sorby published an important paper on this subject in vol. vi. of the 'Monthly Microscopical Journal.' Fresh blood gives two dark bands in the green part of the spectrum, as is shown at F. This colouring matter is hæmoglobin, and the series of changes produced by several reagents may be observed there. A small quantity of fresh blood diluted with a little water should be inserted in the tube shown in Fig. 218, and, examined upon the stage of the microscope, the two bands in the green will be plainly perceived. Next add a trace of ammonia, a small quantity of Rochelle salt (double tartrate of potash and soda), and a particle of the protosulphate of iron and

ammonia (ferrous amm. sulphate), and examine again. The two bands will gradually fade and be replaced by a fainter, though broad and single, band, as shown in G. If this be allowed to oxidise in the air, the two original bands of hæmoglobin can be seen again, and on adding a little citric acid the spectrum becomes changed to that seen in H. The citric acid has changed the hæmoglobin into hæmatin, and this latter can be further deoxidised by contact with ferrous sulphate, when it produces the spectrum shown at I.

Other spectroscopic changes can be induced in blood, but for these we must refer the reader to the tenth Report of the medical officer of the Privy Council, 1867.

In all work such as this, it is advantageous to be able to throw up a second spectrum alongside the first for comparison; and in all good instruments this is provided for by fixing a small prism across half the slit, which receives its light from the *side* of the tube, dispersing it upwards to the eye.

For registering the position of the absorption bands two methods have been devised, (1) the quartz scale of Mr. Sorby, and (2) the bright spot micrometer of Mr. Browning. The first is constructed of two Nicol prisms, with an interposed plate of quartz, about ·043 of an inch in thickness, cut parallel to the principal axis of the crystal. This produces twelve interference lines or black bands in the spectrum, and is adjusted so that the sodium line of the solar spectrum is exactly midway between the third and fourth band, reckoning from the red end. This adjustment is performed by varying the thickness of the quartz plate, by no means an easy operation, as a very slight difference in thickness upsets the scale completely.

The Sorby quartz scale is small and convenient, and is shown at Fig. 220.

The use of this scale monopolises one of the two spectra,

so that in many cases Mr. Browning's bright spot micrometer is decidedly preferable, as it does not preclude the use of either. The lateral piece AA is attached to the upper part of the prism tube, as shown in Fig. 221. At its outer extremity is an oblong box containing a glass plate, upon which is photographed a minute biconvex spot—bright on a dark ground. This is moved vertically by means of a screw connected with the graduated head M, the spot is illuminated by the small plane metallic mirror R, and its image formed by the lens C is capable of being adjusted for focus by the milled collar B. This image is reflected to the eye of the observer from the face of the upper prism, inclined at an angle of 45°, and is viewed along with the two spectra between which it appears to pass when the milled head M is turned. In the construction of maps, notice how many revolutions of the milled head M will carry the bright spot through the whole length of the spectrum, which will vary according to the dispersive power of the prisms.

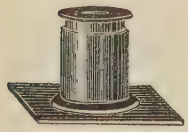


FIG. 220.

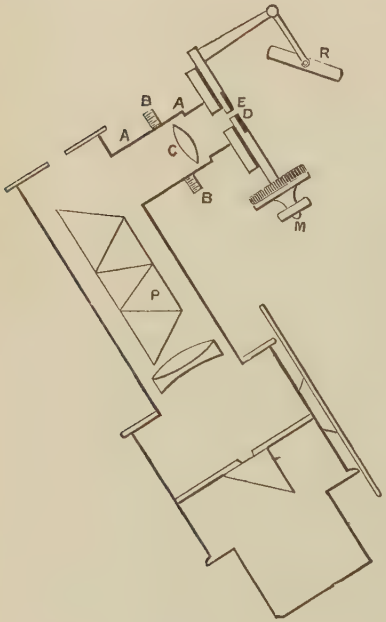


FIG. 221.

A centimetre will be found a convenient measure to represent one revolution: the millimetre will then equal ten of the smallest divisions of the milled head, or $\frac{1}{10}$ of a revolution; these smallest divisions, or thousandths, need not be regarded, as without them the spectrum will be divided with sufficient accuracy. Next measure the distances between the Frauenhofer lines. This should be done with great care, as when once determined it will be constant. The lines A and *a* need not be measured, as they are in the dark part of the red, and are only to be seen in an extremely bright light, so will hardly be required in practice. The same remark will apply to the line H. The spectrum to be measured will therefore extend from about B to a little beyond G.

Let this scale be drawn on cardboard and preserved for reference. Now measure the position of the dark bands in any absorption spectra, taking care for this purpose to use lamplight, as daylight will give, of course, the Frauenhofer lines, and tend to confuse the spectrum. If the few lines occurring in most absorption spectra be now drawn to the same scale as the solar spectrum, on placing the scales side by side, a glance will show the exact position of the bands in the spectrum relatively to the Frauenhofer lines, which thus treated form a natural and unchangeable scale. But for purposes of comparison it will be found sufficient to compare the two lists of numbers representing the micrometric measures, simply exchanging copies of the scale of Frauenhofer lines, or the numbers representing them will enable observers at a distance from each other to compare their results, or even to work simultaneously on the same subject.

Owing to the sliding and other fittings between the prisms and the slit, the micrometer cannot be depended on for pointing to the position of a line by setting it to the

number recorded on the scale, but is only to be used for measuring between lines, and for this purpose it may be trusted.

Dr. Lawson has suggested that the great advantage of this contrivance is that it does not monopolise one of the two spectra, as with the use of the quartz scale; for in describing two spectra, only slightly differing from each other, it may be used at once to determine the difference between them.

In concluding this chapter it may be as well to give a short list of substances showing well-defined absorption bands, such as may be of use to the student upon commencing to use the instrument.

Nitrate of didymium.
Oxalate of ,,
Oxide of ,, (blowpipe-bead).
Chloride of cobalt (in alcohol).
,, ,, (in chloride of calcium).
,, ,, (a crystal).
Cobalt glass (blue).
Sulphate of uranium.
Acetate of ,,
Oxalate of chromium and soda.
Sulphate of chromium.
Permanganate of potash.
Ruby glass (copper).
Ammonio-sulphate of copper.
The aniline colours.
The naphthalene colours.
Indigo sulphate.
Carmine.
Litmus (blue and red).
The colouring matters of plant petals.

CHAPTER XII.

STAINING AND INJECTING.

In many instances it is necessary to stain a preparation—*not merely to form a pretty object*—let this be understood at the outset; but to show certain details of formation not easily discerned in an unstained specimen. When aniline blue and magenta are used for the double staining of vegetable tissues, such as the section of a Burdock stem, Mr. Barrett tells us the different parts are stained as follows:—

Pith	Very pale magenta.
Cellular tissue	Deep magenta.
Spiral vessels of medullary sheath	Deep blue.
Pitted vessels	Blue.
Cambium	Deep blue.
Liber cells	Dark magenta.
Laticiferous vessels	Deep blue.
Cuticle parenchyma	Pale blue.
Epidermis	Deep blue.
Hairs	Pale magenta.

Mr. Stiles, in a paper to 'The Northern Microscopist,' makes the following remarks upon this subject:—

Stems of all kinds should, if possible, be cut when fresh. If they cannot be obtained in this state, they may, previous to cutting, be soaked in cold or tepid water, or in a mixture of equal volumes of spirit of wine, glycerine, and water. Fresh stems or roots can be preserved in this medium for almost any length of time, and will remain in excellent condition for the section machine.

Logwood Staining.—Wood sections require bleaching

before being stained. The bleaching solution is made by mixing ¼ oz. of chloride of lime with a pint of water, shaking occasionally for an hour, and after allowing the sediment to subside, decanting the clear solution. The process of bleaching should be carefully watched and stopped when complete. Tissues vary so much in colour and density that no fixed time can be given for bleaching them. Very thorough washing is necessary. The elimination of the chlorine will be much facilitated by placing the sections, after removal from the bleaching liquid, in a solution of sulphite of soda (1 drachm to 4 oz. of water) for an hour, then washing the sections by soaking them for at least six or eight hours in water, changing occasionally, and finishing with distilled water. If they are not to be stained at once, they should be preserved in water containing 20 per cent. of alcohol, as when kept in water only, in the course of two or three days they become covered with a peculiar fungus growth. At this stage all air-bubbles should be removed from the tissue. This is conveniently done by placing the sections in dilute alcohol, putting them under the receiver of an air-pump, Fig. 222, and exhausting the latter, repeating the pumping occasionally so long as air-bubbles are given off. For this purpose a small tube bottle is employed about 1½ inch long, and a receiver just large enough to hold it, the process being thus rendered a rapid one.

Where it is required to uniformly stain the section in order to render prominent the more delicate cell walls, logwood answers exceedingly well and is very permanent.

LOGWOOD SOLUTION.

Logwood, in coarse powder	2 oz.
Distilled water	10 oz.

Boil for half an hour in a glass beaker, replacing what is lost by evaporation; strain, and to each ounce of liquid

when cold add 60 grains of alum and one drachm of alcohol, rub well together, filter through paper, and preserve in a stoppered bottle.

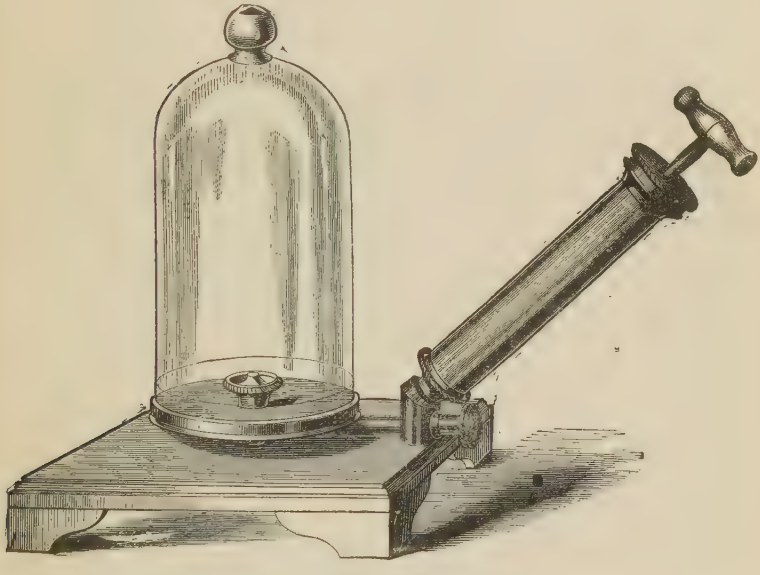


FIG. 222.

STAINING PROCESS.—Make a filtering cone by twice folding a piece of filtering paper about 1½ inch diameter; support this in the neck of a small beaker or tube, and filter through it about ten drops of the above liquid, add thirty drops of distilled water, place the sections in the mixture for five minutes (more or less), pour off the stain, wash once or twice in distilled water, then soak for half an hour in a solution of alum (20 grains to the ounce), remove this, wash well with distilled water, and preserve in alcohol so as to be ready for mounting.

The logwood solution prepared in this way gives more satisfactory results than when made as usually recom-

mended from extract of logwood, the latter being a very variable article.

For double staining, which is more generally useful than the method just described, many formulæ have been given, the process having originated with Mr. G. D. Beatty, of Baltimore, U. S. A.

Perhaps the best paper in this country upon the subject is that which appeared in the pages of 'Science-Gossip,' for January 1880; but even the process therein described is in several points open to modification and improvement.

Mr. A. J. Doherty, in a paper to 'The Northern Microscopist,'* describes at some length the method of staining in carmine and green. The art consists of five stages or processes—1, decolorising the sections; 2, washing the same; 3, preparing for staining; 4, staining in carmine; 5, staining in green.

After bleaching and thoroughly washing to eliminate all the chemicals, as already described, in order to obtain deep colours, the sections must be steeped in a mordant composed of a 10 per cent. solution of alum and water for twenty-four hours, at the end of which time they will be ready to be placed in the first staining fluid, the formula for which is as follows:—

Carmine	15 grains.
Ammonia	15 grains.
Water	2 oz.

The carmine is to be dissolved in the ammonia over the flame of a spirit-lamp, the water added next, and the fluid filtered before it is used.

Immerse the sections in this stain for six or eight hours, then take them out, and wash them in not more than two

* June 1881.

changes of water, and finally transfer them to the green stain, for which take—

Aniline green	5 grains.
Absolute alcohol	1 oz.

Dissolve in a test tube, using a slight heat only, to avoid any unpleasant mishap, and filter before using. After a three hours' soaking in the above, the staining will be completed, and the sections should be mounted without delay, after having washed the superfluous colour away with methylated spirit. The method of mounting these will be given later on.

*Staining in Picro-carmine.**—Picro-carmine is the most truly selective of any double stain yet employed. A special modification of it is required for wood sections as follows:—

PICRO-CARMINE SOLUTION.

Carmine (finest)	2 grains.
Liquid ammonia (sp. gr. ·960)	½ drachm.
Distilled water	1 oz.

Put the carmine in a 2-ounce stoppered bottle, pour in the liquid ammonia, and shake occasionally until dissolved, then add the water.

Picric acid	8 grains.
Alcohol	1 oz.

Dissolve in a test-tube with a gentle heat, then mix with the solution of carmine.

STAINING PROCESS.—Place the sections in 50 per cent. alcohol for one hour, then treat with the recently filtered staining solution until the desired effect is produced (usually from half to two or three hours), remove the dye, wash quickly three or four times with alcohol 50 per cent., then soak in an alcoholic solution of picrate of ammonia,

* Mr. M. H. Stiles in 'The Northern Microscopist,' July 1881.

changing this at the expiration of an hour, and allowing the sections to remain in the second solution for about the same period.

The logwood stained sections, after being well washed, are soaked in alcohol for an hour, then removed to oil of cajeput, and allowed to remain in this for a couple of hours; at the end of this time transfer them to oil of turpentine. In less than an hour they will be ready for mounting in balsam or dammar. The sections should not be allowed to remain long in the turpentine or else they become brittle.

In the case of picro-carmine stained sections they should be removed from the alcoholic solution of picrate of ammonia into alcohol for about a minute, then into oil of cajeput.

The object of employing an alcoholic solution of picrate of ammonia is to avoid the loss of colour which attends the use of alcohol only, the yellow stain of picric acid being readily removed from the tissue by that liquid. Picrate of ammonia may be easily made by adding a slight excess of liquid ammonia to a solution of picric acid, and evaporating the mixture to dryness at a gentle heat.* The residue is dissolved in alcohol and filtered.

Wood sections stained in picro-carmine are very beautiful and permanent. The staining being done at *one* operation, and the colours being remarkably selective, there is an absence of secondary tints, as in the case of most other double stains, especially where one tint is partially washed out to make way for another.

Regarding permanence, some stained sections mounted nearly five years ago appear to have retained their brilliancy unimpaired.

* This salt had better be purchased by the student, as the picrates explode violently when over-heated.

In place of alcohol, methylated spirit may be used if desired.

The chromo-lithograph forming the frontispiece to this work, illustrates very clearly the value of the foregoing methods. The four outside figures are specimens of double-staining in carmine and green, some of the admirable specimens of Mr. Ward, of Manchester, while the centre figure is a representation of the slide sent by Mr. Stiles, of Doncaster, to illustrate his paper on picro-carmine staining in 'The Northern Microscopist.'

Histological (animal) staining differs somewhat from the foregoing, many processes having been devised to strikingly differentiate the tissues. In 1876, Dr. Elizabeth Hoggan described a process with iron and pyrogallic acid as follows:*—

"The colouring agents required are a one or two per cent. solution of perchloride of iron in alcohol, and an alcoholic solution of pyrogallic acid of similar strength."

The section or membrane to be stained is first treated for a short time with alcohol, the iron solution filtered upon it, and then poured off after a couple of minutes. The pyrogallic solution is then filtered upon it, and when the desired depth of staining has been obtained, the tissue is washed and mounted in any of the usual ways.

The nuclei and nucleoli are by this means coloured black, and the cell-substance coloured more or less. A bluish tint may be imparted by washing in alkaline water or in water highly charged with carbonate of lime.

Osmic acid has been frequently recommended for staining animal tissues black, but it is doubtful whether anything is gained by its use. Dr. G. Brösicke, of Berlin, advises a mixture of osmic and oxalic acids for this

* 'Journal of the Quekett Club,' iv. p. 180.

purpose. Thin sections of animal tissues are placed in a one per cent. osmic acid solution, and then carefully washed to remove all superfluous acid. They must then be immersed for about twenty-four hours in a solution of one part of oxalic acid to fifteen of water, when after washing they are ready for examination.

The oxalic acid produces darker or lighter shades (carmine and Burgundy tints) in proportion to the length of time the section has been immersed in osmic acid, but if the tissue has once become blackened, the oxalic solution is powerless to redden it afterwards.

Chloride of gold solution has been employed for colouring nerve-fibres; a solution in water of one or two grains to the ounce is generally used. The section should be soaked in the solution until it has acquired a straw-coloured tinge, it is then to be washed and placed in a one per cent. solution of acetic acid. In the light the nerve-fibres become coloured a blue or violet tinge in a few hours. Nitrate of silver is a very important agent for the staining of animal tissues. Dr. Lionel Beale writes, "A weak solution may be imbibed by delicate tubes, and part being precipitated in the tube, perhaps as a chloride, or in combination with some albuminous material, subsequently becomes decomposed by the action of light."

Recklinghausen, who has employed this plan with much success, uses a very dilute solution, made by dissolving one grain of nitrate of silver in from one to two ounces of distilled water.

In 1863, Dr. Roberts, of Manchester, used a solution of tannin and magenta for staining the red human blood-corpuscles, and this is a subject which still requires investigation.

Dr. Beale's carmine fluid for staining all forms of *bioplasm* of living things is made as follows:—

Carmine	10 grains.
Liquor ammoniæ	½ drachm.
Glycerine	2 oz.
Distilled water	2 oz.
Alcohol	½ oz.

The carmine is to be dissolved in the ammonia, boiled for a few seconds, and set aside for an hour; the glycerine, water, and alcohol may then be added, and the whole allowed to stand until thoroughly settled, when the clear fluid is to be decanted and kept for use. Dr. Beale tells us that if the solution be very alkaline, the colouring will be too intense, and much of the soft tissue round the bioplasm will be destroyed by the action of the ammonia; if, on the other hand, the solution is neutral, the uniform staining of tissue and bioplasm may result. In some cases the fluid must be diluted with alcohol, water, or glycerine, in order to get the best results; and Dr. Beale observes that the process should not be hastily condemned without trying the effect of modifying the quantities of the various constituents.

There are many other processes to be found scattered through the pages of microscopical literature, but space compels us to close the subject here.

The process of injecting should now claim our attention, though it is feared that actual practice with the syringe will be found of far more use than any written instructions can ever be.

There are two methods of making injections, opaque and transparent; in the former the colouring matters, such as vermilion, chromate of lead, white lead, or Prussian blue, are well mixed with size made of gelatine, of such a strength that it will form a firm jelly on cooling. To make the size, soak one ounce of gelatine in a pint of water overnight, and dissolve with heat in the morning.

The colouring matters must be mixed up in a mortar

with the size, poured into a tin vessel placed in hot water, and strained through fine muslin into another, just before it is required to be used. It must not be forgotten that when size is used, the preparation to be injected must be steeped in hot water, and kept warm until the operation is finished.

Opaque injections in various colours often form very instructive objects. An injection of the liver illustrates this very well: the artery may be injected with vermilion, the portal vein with white lead, the duct with Prussian blue, and the hepatic vein with lake. A section of an injected kidney, with Prussian blue and lake, is also a pleasing slide when well performed.

After all, opaque injections have but a limited scientific value, and therefore the student is advised to utilise his energies upon the transparent class, as they can be examined with all powers, and when preserved in fluids the most delicate structures retain their integrity.

The Prussian blue fluid for transparent injections is made as follows (Beale):—

Glycerine	1 oz.
Alcohol	1 oz.
Ferrocyanide of potassium	12 grains.
Tinct. ferri perchlor.	1 drachm.
Water	4 oz.

The ferrocyanide is to be dissolved in an ounce of the water and the glycerine added, while the *Tinct. ferri perchlor.* is separately mixed with another ounce of the water. These two solutions should now be mixed very gradually, and well shaken, the iron being added to the ferrocyanide; the mixture of spirit with the remainder of the water must now be added very gradually, when the fluid is ready for use.

Dr. Beale's carmine fluid is made in the following manner:—

Liq. ammoniæ	5 drops.
Carmine	5 grains.
Glycerine	1½ oz.
Acetic acid	8 drops.
Glycerine	1 oz.
Alcohol	2 drachms.
Water	6 drachms.

Dissolve the carmine in a few drops of water, and the liquor ammoniæ at a gentle heat, and when dissolved add half an ounce of the glycerine. Shake well, and then add gradually half an ounce of the glycerine in which the acetic acid has been dissolved, well shaking the bottle between each addition. If not decidedly acid to litmus paper, a few more drops of acid must be mixed with the remainder of the glycerine, and added as before. Lastly, add the alcohol and water very gradually, well shaking until the whole is well mixed.

The author does not intend to enter further into the details of this branch of microscopy; with the foregoing solutions the student will be enabled to learn the elements of the art, and if afterwards he has a desire to proceed further, he will find full instructions for many fluids in 'Das Mikroskop,' by Dr. Frey, or in the admirable treatise of Dr. Lionel Beale.

The operation of Injecting.—The various steps in the process of injecting are very similar, whether the object be treated for the opaque or transparent methods, though in the former instance it must be placed in warm water until the temperature has been sufficiently raised.

The necessary apparatus will be found as follows:—

A syringe with pipes and stopcocks.
Conical corks.
Dissecting knives (Fig. 153).
Dissecting scissors (Figs. 154–56).
Forceps (Fig. 158).
Bull-nose forceps.
Wash bottle (Fig. 223).

The first operation is that of finding the vessel we wish to inject, such as the artery of the kidney, or the hepatic vein of the liver. An opening being made, one of the small pipes of the syringe is thrust in and tied round with a piece of waxed silk, so that there may be no danger of withdrawing the pipe before the conclusion of the operation. All the vessels communicating with the one in which the pipe is connected must now be tied with silk to prevent the escape of the injecting fluid, and several pairs of bull-nose forceps should be at hand for the purpose of closing any which may by accident have been left open.

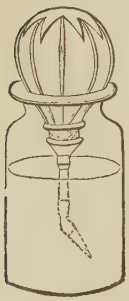


FIG. 223.

Before the pipe is inserted a little of the injecting fluid should be drawn up to fill it. When all is ready, the syringe should be filled and emptied a few times with the fluid in order to get rid of all air, especially below the stopcock, and fitted to the pipe already in the preparation. With a firm pressure the syringe must be about three-fourths emptied, again refilled and injected until the operation is complete.

When size is employed, after having tied up the vessel where the pipe was inserted, the whole should be left in cold water for five or six hours, in order that the size may thoroughly set, after which sections may be cut as directed in Chap. VIII.

Injected specimens may be preserved in glycerine or in a mixture of equal parts of alcohol and water, until required for examination ; carbolic acid water is also useful for this purpose.

CHAPTER XIII.

THE PREPARATION AND MOUNTING OF OBJECTS.

We have now described most of the processes incidental to microscopical research, and which should be applied to specimens in order to gain some insight into their peculiarities or their structure. No doubt many students will think that we have delayed unnecessarily the preparation of objects for mounting, and the subsequent process whereby they may become permanent objects of interest, and that we have taken pains to describe in detail processes much more difficult than mounting.

We do not think this. It may be thought an easy operation to place a piece of sea-weed in Canada balsam, and cover it with a circle of thin glass; or to soak an insect in potash, to squeeze out the internal organs, finally mounting in balsam and benzol; but this is not the style or class of mounting which we wish to see become general.

Dr. Pelletan, in a letter to the 'Journal de Micrographie' (iii., No. 3, p. 139), speaks very strongly of the scientific value of microscopical preparations. In speaking of the method of mounting insects he says: "Others, more ingenious, mount large insects or immense spiders entire after having emptied them of their contents, and these preparations have really a magnificent appearance. But alas! the integument is all that has been preserved, and the little that remains of the internal organs is represented by a uniform transparent mass, in which the microscopist finds nothing to study." He also adds further on: "I would

not say that all preparations which I call trivial are useless; most certainly not. If they are not satisfactory to *savants* they interest *amateurs*, and they teach many things that otherwise would not have been known. They are also useful in England, where they are sold in large numbers, because among our neighbours the microscope is more used for amusement and as an object of luxury than for working purposes."

We cannot fully endorse the opinions of Dr. Pelletan, though it would be more satisfactory to find an improvement in the general mounting of objects; and to prepare one or two slides of a subject *well* would be of far more value than mounting a host of second-rate ones for *exchanges;* and further, we would like to see far more experiments upon subjects mounted in different media than exists at present, and what with interchange of opinions upon this subject at our various microscopical society meetings, we could scarcely fail in rendering our cabinets more interesting and our preparations more permanent.

The mounting of objects and their preparation for this purpose is by no means an easy operation, especially if our slides are to be *permanent* and of scientific interest. The conditions are ever varying, and it requires a good knowledge of the properties of the various substances generally used in this branch in order to know beforehand what their actions will be on this or that object or portion of it. Even a process which would do well for some particular class of objects is often found to fail with some of them. If, for instance, the marine alga *Bangia fusco-purpurea* be mounted in balsam and benzol it will represent it in its natural condition, while if we try to preserve *Dasya coccinea* in the same manner we shall fail miserably. In other instances we may find many algæ which may be successfully mounted in Deane's medium, while if we try to put up the first-

named algæ in it we get nothing but a swollen tube containing swollen endochrome. Thus it is that many objects are spoiled by the lack of knowledge of various preparers, and also by the fact that some who should possess this knowledge perform their operations so rapidly that it is impossible they can produce uniform and permanent work. And what do we see when we cast our eyes over the contents of various cabinets? Drawer after drawer is scanned, and if the third of their contents are passable, *from a scientific point of view*, the possessor may be congratulated. What can be done to get microscopy out of this groove? Slower work, more time in preparing, more care, the rejection of all middling or bad slides, the study of the object before proceeding to mount it, mounting but few slides, and last and not least, the careful study of the effects of the various varnishes, cements, and reagents upon each other, and upon the various objects they are intended to preserve.

For the preparation and preservation of objects for the microscope certain pieces of apparatus are either necessary or useful, and although many makeshifts can be employed, we give illustrations of the various instruments most generally used.

Objects are generally mounted upon glass slides, or "slips," as they are sometimes called, which measure three inches in length by one in breadth, and of various thicknesses. They are sold either with rough edges or ground edges as may be required, but there is so little difference in price between the two varieties—the latter possessing so many advantages—that the student is strongly advised to purchase *ground edges* only.

The ordinary flatted crown slips are the cheapest, but should never be used for fine work, those of plate glass being preferable, and even this should be selected if required for mounting objects requiring delicate attention to illumi-

nation. All kinds are generally fairly clean when received from the dealer, but must, nevertheless, be beautifully polished before any object can be mounted upon them. The thorough cleansing of the glass is an important step, as the slightest film of grease is a preventive of perfect adhesion of any varnish or cement which may be subsequently used. Ammonia has the power of converting grease into soap, and spirits of wine will dissolve the two, and as some kinds of dirt require friction to remove, the following is perhaps the best formula for a mixture for cleaning slides :—

Liquid ammonia (sp. gr. ·880)	10 drops.
Methylated spirit	2 oz.
Water	½ oz.
Rouge, sufficient to make a thin cream.	

Rose-wood slips, also 3 inches long by 1 broad, are also used for mounting objects, either with sunk cells or with holes bored right through ; but the student will find, perhaps only after a valuable series has been spoiled, that wooden slides possess no advantages over those of glass, and have very many objectionable qualities.

FIG. 224.

In mounting objects, a slide-centerer should be employed, in order to make the finish as presentable as possible, as it will generally be found that a slovenly finish means a bad preparation, though this is by no means *always* the case. The most simple form of centerer is shown in Fig. 224,

which is simply a piece of very stiff card, upon which the lines and circles can be drawn.

Another form of centerer for slides was illustrated in 'Science-Gossip' for 1879, from which the accompanying sketch is taken (Fig. 225); the device is so simple that it needs no description.

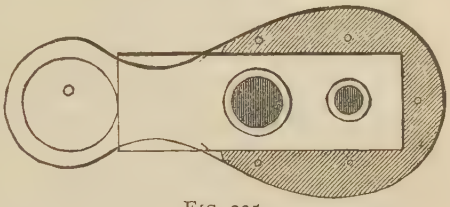


FIG. 225.

The object is nearly always covered with thin glass, either squares or circles, and is sold by the ounce in sizes for circles, increasing by eighths of an inch. It is of importance that for high powers used dry the cover-glass should be of uniform and great thinness. When purchased they should be selected according to thickness, and each variety used for its special purpose. The thickness may be ascertained in various ways. In one, the cover may be held in the stage forceps edgeways, and measured by observation with the micrometer eye-piece shown at Fig. 202.

Another method is by the lever of contact as made by Messrs. Ross and Co., or the cover-glass measurer devised by M. Schönemann, called the "measuring wedge," and shown in Figs. 226 and 227.

FIG. 226.

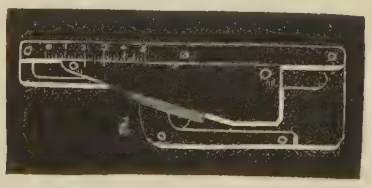


FIG. 227.

In order to measure the thickness of a cover-glass the instrument should be free from dust, and the wedge drawn back until the cover can be placed between the wedge and

the piece *d*. The wedge is then closed until the motion is stayed by the glass, when the number of the divisions indicating the thickness can be read off.

None but the very thinnest glass can be used with the dry $\frac{1}{25}$ and $\frac{1}{50}$ objectives; it is much more expensive and difficult to cut than ordinary cover-glass.

Cells and Cell-making.—Many objects requiring to be permanently mounted are of such a thickness that the thin cover needs some support at its edges, or again others are better preserved when mounted in fluids, and for these pur-

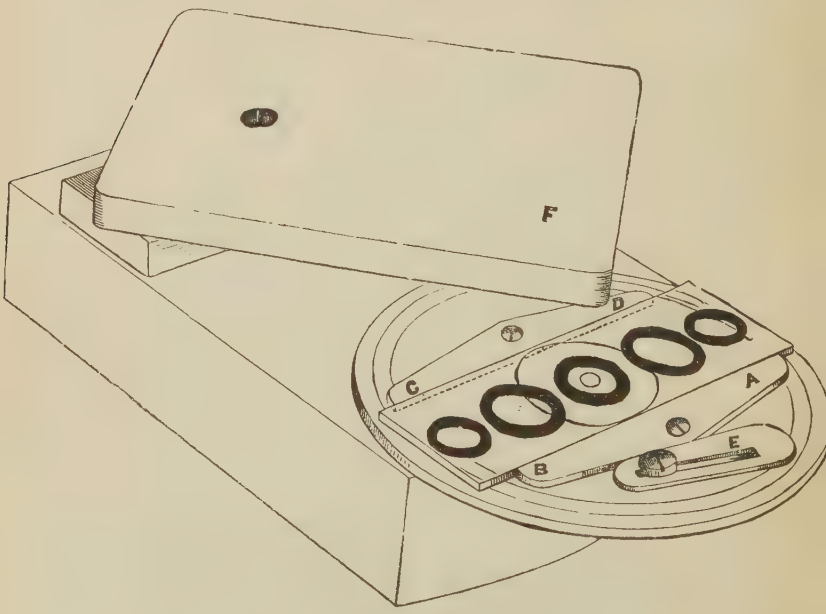


FIG. 228.

poses cells are employed. These are of several kinds, but may be conveniently divided into three classes, varnish cells, solid, and built-up cells.

The cell-making machine was originally devised by Mr. Shadbolt, as a simple brass circular plate about three inches in diameter, upon the surface of which was placed a pair of

springs for the purpose of holding down the glass slip ; it has been the parent of several forms supposed to be of great benefit to the microscopist. In order that the slides may be easily centered, the table is usually engraved with a series of circles ; but it is readily seen that such centering can only be approximate.

In order to centre the slides accurately in one direction, that of width, Zentmayer introduced the simple device of fixing a couple of pins equidistant from the centre and at opposite sides of the table, the slide being so arranged that it touched both of these pins. This centres for the width, and in length this is accomplished by a series of circles near the edge of the table, the operator making the adjustment from inspection of these.

In 1870 Dr. Matthews devised a turntable (Fig. 228) to accurately centre slides in the direction of their width, and this possessed the further advantage that no springs or other portions of the table rose above the slide, to catch the fingers or brush, during its revolutions ; and in the next year Mr. J. B. Spencer, in a communication to 'Science-Gossip,' showed how this might be made in hard wood by the microscopist himself.

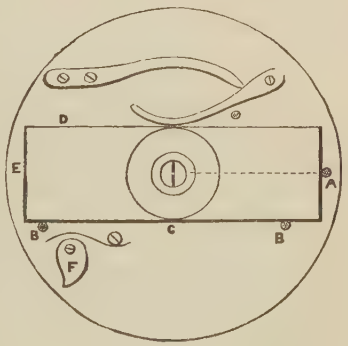


FIG. 229.

In 'Science-Gossip' for 1874 Mr. Bridgman described and illustrated a form of turntable (Fig. 229) which, though not self-centering, enabled a slide to be always placed in the same position upon it, so far as the centre of rotation went. In 1875 Mr. C. F. Cox, of New York, devised a self-centering turntable, consisting of the circular revolving plate, in which was cut a slot in the direction of its diameter, and in this were

moved, by means of a right-handed and left-handed screw, a pair of clips which gripped the opposite and extreme corners of the slip, as shown in Fig. 230.

It will thus be seen that so long as the edges of the slip are at right angles to each other the centering must be absolutely accurate, but not otherwise.

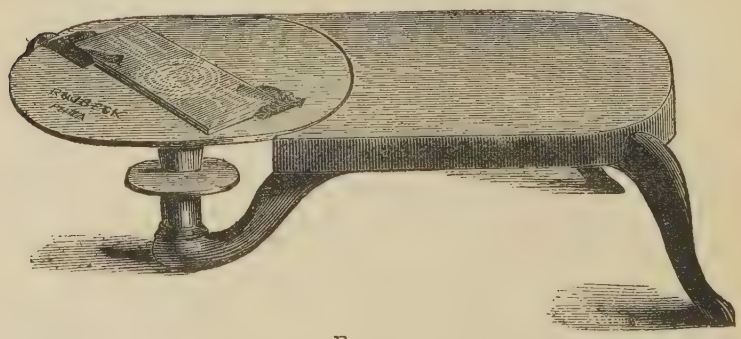


Fig. 230.

On the introduction of this machine several operators objected to the method of holding the slip, and Kinné soon afterwards introduced his modification (Fig. 231), in which the two corner clips were drawn together and made to grip the glass slip by means of an indiarubber band or spiral spring.

In 1876 a notice appeared that Mr. Charles Butterworth, of Shaw, near Oldham, exhibited at the annual soirée of the Oldham Microscopical Society a turntable capable of making cells of either circular or elliptical form; and also by its aid a thin cover-glass could be held in position on a cell, whilst the various rings of cement or varnish could be put on. It is constructed upon the principle of the "oval chuck," and so enables either circles or ovals to be traced with ease—moreover, it may also be used for cutting thin glass covers, either oval or circular, as well as for general mounting purposes.

A turntable of exactly similar make is now sold by Messrs. Armstrong, of Manchester; it is shown in Fig. 232.

FIG. 231.

Mr. Sidle exhibited, in 1878, at the Microscopical Congress at Indianapolis, a turntable, which has since gone by

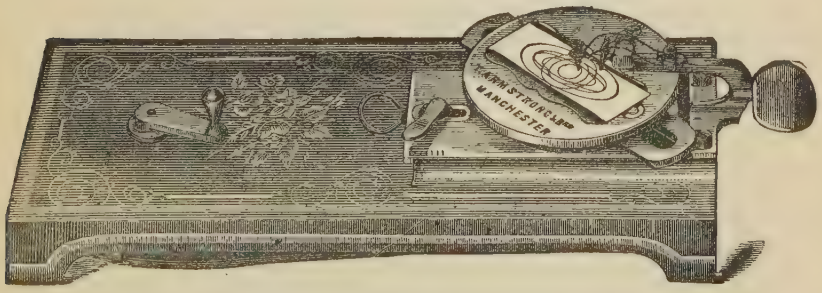


FIG. 232.

the name of the "Congress" turntable. It may be seen in Fig. 233.

Into the upper surface of the rotating plate, diametrically

opposite and equidistant from the centre, two circular plates or discs, one inch in diameter, are set, their surfaces flush with that of the large plate. Pivots from the two discs project through the plate, and each carries upon the

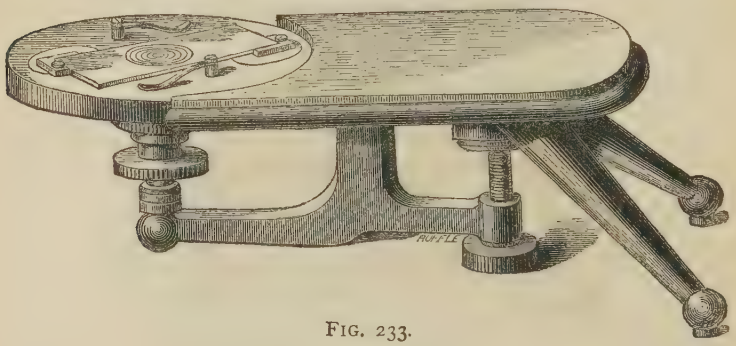


FIG. 233.

lower side of the plate a toothed wheel. A hollow sleeve, rotating free upon the stem of the table, carries a third and larger wheel, which gears into the two others, and thereby gives rotation to the discs in the top of the plate.

Near the opposite edges of the two discs, the angular jaws which hold opposite corners of the slide are pivoted (as in Cox's and other forms of tables), and it will be seen that by giving rotation to the central wheel, under the plate, the jaws may be made to approach or recede at pleasure.

A coiled steel spring, concealed within the hollow sleeve, serves to close the jaws, while the single motion of a milled head upon the sleeve opens them to their full extent.

It may be also seen that although the jaws do not approach in a straight line, yet, when properly adjusted, a line joining the pivots of the jaws will cut the centre of the plate, whatever the position of the jaws; and they being always equidistant from the centre, it follows that

the slide, when clasped between them, must be perfectly centered.

In 1879, Mr. Rolfe re-invented Kinné's turntable, and described the same before the members of the Quekett Club, adding at the same time an idea of his own, which was quite novel, and is illustrated in Fig. 234.

FIG. 234.

The year 1880 saw the introduction of two turntables, the first by Mr. Dunning (Fig. 235), which has since been made by Mr. Swift, and the second introduced by Dr. Matthews. Mr. Dunning's turntable will take any slides up to two inches in width, and also serves for retouching slides, the circles upon which are not truly central. Dr. Matthews' last production is to be found fully described in the 'Journal of the Royal Microscopical Society,' and as it only consists of a method of driving the table, it will be of but little use mentioning it here in detail.

Another good form of self-centering turntable has lately

been introduced by Mr. H. P. Aylward, of Manchester, and may be said the simplest, least expensive, and best self-centering turntable extant.

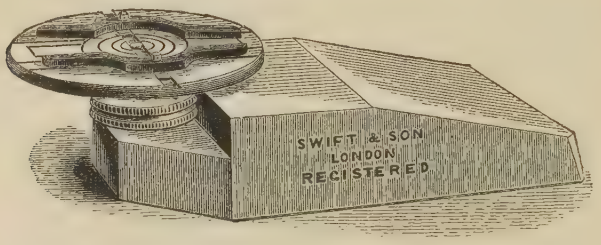


FIG. 235.

The author, from actual experience, can recommend it as being *the* turntable for all the wants of the microscopist. Mr. Aylward calls it the "Concentric." It essentially consists of two plates, the inner revolving on a pivot, whilst the outer revolves concentrically on the inner, a few small pins being so arranged that by a single turn of the outer ring they firmly grasp the glass slide, and cause its centre to exactly coincide with the centre of the turntable, whilst a simple reverse movement instantly liberates it. This turntable answers for slides of various widths, from 1 inch to 2½ inches, it is strongly made, and well finished, besides which loose springs are supplied, fitting into corresponding holes in the turntable, convenient for making rings in any other position than that of the true centre of the slide.

This instrument is shown in Fig. 236, the letters being explained as follows:—

A, ordinary wood block with steel pivot, on which the brass table revolves. B, two brass springs which fit into holes in the table, and may be used when the slide is required to be out of centre; when not in use they fit into holes in the wood, as shown above. D, revolving

table, with milled wheel below for rotation. H, brass annulus or ring revolving concentrically on the table D; on the ring H are screwed two conically headed pins, J J, $3\frac{3}{32}$ of an inch apart exact, to allow the 3 × 1 slip to be placed diagonally between them. F F, two similar pins in

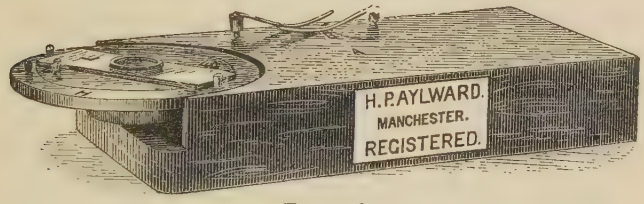


FIG. 236.

plate D, so placed, that upon revolving the ring H they, in conjunction with the pins J J, firmly grasp the opposite corners of the glass slip, and cause the centre to coincide with the centre of the table D. I, brass pin for more easily revolving the ring H, for securing and liberating the glass slip, which is done by moving the ring H in the opposite direction.

Let us now turn our attention to the practical operation of cell-making. To commence with varnish cells—place the slide upon which the cell is to be made between the pins upon Aylward's turntable, and make three rings thereon (of the diameter the cell is to be when finished) with a writing diamond: this operation is to roughen the slip, and cause more perfect adhesion of the varnish. Now take up a good brushful of varnish, and spinning the table round, deliver it where the rings have been cut, in such a manner that it stands up like a wall, and does not spread itself more than is necessary over the slide. So soon as this layer is dry and perfectly hard, another layer may be put on, and the process repeated until the cell has acquired sufficient depth. The cell now requires drying or baking

at a gentle heat over a long period, and this can be easily accomplished in the hot-air chamber shown in Fig. 237.

This piece of apparatus is not absolutely necessary, though it is of much assistance. By means of a spirit-lamp or gas-burner almost any temperature can be maintained for lengthened periods, a thermometer inserted into the air space serving to measure the degree of heat; it serves many useful purposes, and is certainly better and more uniform than the "cool oven" so often recommended.

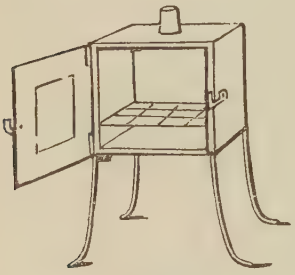


FIG. 237.

The varnish of which a cell is constructed must vary according to the nature of the medium it is to hold; for objects mounted dry and not very thick, a brown varnish cell is all that is required (see receipt, Chapter XIV.), or it may be made of asphaltum, marine glue, or gold-size, at the fancy of the operator.

The gold-size cell requires no baking, the hardening is due to an oxidation process, and wherever used it forms a most reliable cell. Varnish cells should be made some time before they are required, in order that no change takes place after the cover-glass has been put on.

Deeper cells are constructed of glass, ebonite, tin, ivory, brass, and several other substances; the glass slip forming the base of the cell should be roughened as before mentioned, the cell-ring roughened on each side, and then cemented to a glass slip with gold-size or brown varnish.

The student is advised to eschew all paper, cardboard, and wax-cells: varnish is the safest to use for thin cells, and glass, pure tin, or ebonite for the deeper ones, and if

care be taken to roughen the surfaces of contact, the tendency to leak when filled with fluid will be much reduced. A considerable number of cells of various diameters and depths should be made at one sitting, and a stock of old cells always kept on hand. If new ones are used they often turn out unsatisfactorily.

Large cells may be made of four separate walls of glass all ground together to one level, cemented by their corners to the glass slide with marine glue. In using marine glue the surfaces to be united should be heated very hot, the marine glue applied to the edges, and kept firmly pressed together until set. They may then be further heated in the bath, Fig. 237, for twelve hours.

In passing now to the mounting of objects, the reader will see the impossibility of describing how any particular object or class of objects may be successfully mounted; which is the best of the several methods can only be discovered in actual practice, and if the salient points in each method of mounting be described, the student will no doubt learn much that will help him with other subjects.

For this reason the mounting of objects has been divided into three sections—

1. Mounting dry.
2. Mounting in gum resins.
3. Mounting in aqueous media.

DRY MOUNTING.—In treating of dry mounting we may so subdivide the work as to show the isolated operations upon which success depends, and in doing so, the student will see the importance of thoroughly understanding the why and the wherefore of each operation. These may be described as follows:—

Cleaning the Specimens.—This is a section upon which a

moderate-sized volume may be written, as it applies to all objects whether mounted dry, in gum resins, or in aqueous media, and may be simply described as an operation for eliminating matter in the wrong place—dirt. Foreign matters should be eliminated as much as possible, and really when set about in the right way it is not very difficult. When we come to compare the slides of diatoms put up by Cole, Redfern, or Redmayne with many home-mounted slides it may be readily seen what is the effect of a little care on the part of the preparer. Cole's exceedingly clean gatherings, his hand-picked slides, Redfern's single diatom, mounted on a $\frac{1}{4}$-inch cover in the centre of a red circle $\frac{1}{16}$ of an inch in diameter, and Redmayne's diatom slides all deserve imitation.

Let us turn again to the mounting of mosses: how many of the *οἱ πολλοί* are content with placing the specimen in glycerine jelly, dirty as when collected, making no effort at all to divest it of its useless and degrading accompaniment!

It is to be hoped that these few words will act as an incentive to cleanly working; all the requisites are several camel's-hair or sable pencils, one of which should be cut short so that the hair projects but a quarter of an inch beyond the quill-holder. The knives, scissors, forceps, needles and other articles have been already described in the chapter on dissections; it may, however, be necessary to state that each article should be kept for its specific uses, as if knives, scissors, and needles be used for mounting purposes they will soon be out of order for dissections.

Most objects can be cleansed under water with the brushes and needles in the small dissecting troughs shown in Fig. 152; these can be used under a watchmaker's eye-glass or on the stage of the microscope under a half-inch, with erector such as has been already described.

Before being finally finished, every slide should be examined under that power most suited to show its characteristics, and if it is defective in any way it should be discarded and washed off the slip.

The final washing of an object treated with water or any aqueous fluid should be made with distilled water—ordinary water leaves more or less residue, which interferes with the brilliancy of the preparation.

Drying the Specimens.—The moisture can be abstracted from many substances by contact with a fluid such as alcohol in a small corked tube, but in many instances this is not admissible, so that a desiccator becomes necessary. The form used by the author is shown at Fig. 238. It consists of a mahogany base-board, in which a circular groove is turned to admit the bell-jar standing over it. This groove is filled with mercury, which acts as a lute and cuts off all connection with the outer air. Under the bell-jar, and standing upon the base-board, is a vessel containing concentrated sulphuric acid, one of the most powerful absorbers of water we have. A shelf is fixed over the acid, upon which are laid the slides requiring desiccation.

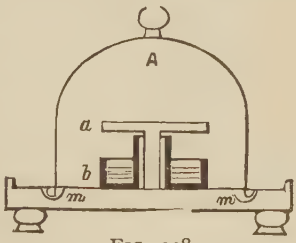


FIG. 238.

Unless the operator is careful and has a safe place to keep the desiccator, he had better choose another pattern, as concentrated sulphuric acid is a fluid not to be spilled with impunity, and quicksilver is an extremely awkward metal to pick up from the floor upon which it may have been dropped accidentally. The sulphuric acid may be replaced by fused chloride of calcium, carbonate of potash, or even by quicklime broken in pieces the size of

hazel nuts; but the apparatus is not then quite so effective or rapid.

The mercury may indeed be dispensed with altogether by the use of a plate of ground-glass in lieu of the grooved base-board, but in this case the bell-jar must have a strong and well-ground welt round its open mouth, perfect adhesion between the two being secured by means of a coating of grease.

There is a very good form of desiccator which may be obtained from almost every dealer in chemical apparatus. The lower portion contains the desiccating material, over which is placed a sheet of perforated zinc, while the cover is ground truly, and fits as a cap or lid upon the lower half. Perhaps this form would suit the majority of our readers, more especially if combined with the use of chloride of calcium as the desiccating agent.

The preparation to be dried is placed upon the shelf (Fig. 238) above the desiccating material, covered with the bell-jar, and left to itself for twelve or twenty-four hours.

In dry-mounting it is absolutely necessary to eliminate *all* the moisture, for many objects mounted in a damp state often become covered in a few months with a dense growth of fungi, or the cover-glass becomes obscured to such an extent that the object is seen as through a fog. This often happens also when a cardboard cell or wooden slide is used.

Let us now proceed to illustrate the foregoing by the process for mounting diatoms given in the 'American Journal of Microscopy,' April 1880.

The process of cleaning diatoms requires time, skill, patience, and personal experience, in addition to what may be learned from others. After trying for a long time to dispose of sand and mud the novice will be more careful in collecting. After an explosion or two, involving the loss of

valuable material, and possibly the destruction of clothing, he will learn that strong acids and other chemicals are not to be handled like water. Experience makes the process safe and comparatively easy, requiring but a few minutes' attention at a time. No one method will apply in all cases, for some gatherings are imbedded in stone, some cemented with lime, which require special attention, while many gatherings require nothing more than a strong heat to destroy the organic matter and leave them ready for mounting.

In recent gatherings, when the diatoms are clean, put them into a bottle containing equal parts of alcohol and water, where they may be kept as long as desired. When ready to transfer them to slides, all that is required with most varieties is to dip a few from the bottle with a pipette, to put them on the thin cover-glass, and after placing the glass on a strip of mica or of tintype, keep the whole at a red heat until the organic matter is destroyed and only the shells remain in white powder.

Another method is to boil for thirty to sixty minutes in strong soapsuds, afterwards washing thoroughly in soft water to get rid of foreign material, such as sand, flocculent matter, &c. On examination of the material, if organic matter be still present, put the mass into a test-tube or other suitable vessel, and, after settling, completely turn off all supernatant water, adding four or five times its bulk of nitric acid, and while boiling throw in small fragments of bichromate of potash to bleach. Some prefer chlorate, but the bichromate is sufficient, and danger of explosion is avoided. When the organic matter has been destroyed, a higher temperature will be required to boil the acid, indicating that no more is needed. Probably five or ten minutes will be sufficient. Wash in rain-water, or that from melted ice, until a drop evaporated on a slide shows no

residue around the edge, leaving a clean slide of diatoms. Never use hard water, for the lime in it will cause all flocculent matter to cohere in masses.

The methods given are all that is required for a large proportion of diatomaceous material so far as disposing of organic matter is concerned. The sand and other indestructible matter must be eliminated by gravity.

Guano, Monterey stone, material containing lime, &c., require harsher treatment and much more time.

Guano should be boiled at least two hours in soft water, or as long as any colouring matter can be turned off; then proceed as in fossil earths.

Stonelike masses must be broken down by boiling in a strong solution of soda crystals. After disintegration wash and boil for twenty to thirty minutes in strong nitric acid, and while yet boiling add about an equal quantity of muriatic acid, continuing the boiling for from twenty to thirty minutes longer. After washing out the acids boil in pure sulphuric acid until the mass becomes inky black, then throw in fragments of bichromate of potash, and continue the boiling until it becomes clean. If, on examination with the microscope, it is found there is much flocculent matter besides the diatoms and sand, it can be removed by boiling for a few seconds in caustic potash, and then turning *almost instantly* into plenty of soft water to destroy the action of the potash. The diatoms are now chemically free from all organic matter, and they may be dried and kept in small phials in powder, or be put into equal parts of alcohol and water, and kept for future separation from sand and other inorganic matter, or we can proceed at once to isolate the diatoms, also to separate into sizes. To do this, put the cleaned diatoms into a small bottle, fill with soft water, filtered, and after shaking thoroughly turn off all that floats after five seconds into a larger bottle. Repeat the process, and after

some five or six repetitions we shall find very little but sand in the first bottle; this we will throw away unless some very large diatoms remain, which can be removed by drying on a slide and picking with a mechanical finger. As soon as the material in the large bottle has settled, turn off the water and return the material to the small bottle and repeat the process, allowing longer time to settle. This process may be repeated five or six times, or as many times as necessary to make the separation satisfactory, allowing more time on each repetition to settle. Another excellent method is used by Christian Febiger, of Wilmington, Del., whose arranged slides have attracted much attention. Strain through No. 18 bolting cloth to obtain large diatoms. The remaining small diatoms and sand must be placed with water in a clock crystal and rotated. The sand will go to the bottom, and the diatoms can be poured off repeatedly until as clean as desired.

It will be impossible to save all the diatoms in the repeated washings. So long as 100 slides can be mounted from a mass not so large as a small pea, be content to save time and patience by losing a tithe of the harvest. Do not be disappointed when you find hardly enough diatoms remaining to make a fair thickness of carpet in your phial, for if clean you will have sufficient for yourself and for several of your friends, even then.

For mounting, always place with a pipette a drop of the fluid containing them upon the cover-glass, and never on the slide. Professor Hamilton L. Smith is the author of the following excellent method. Cut a piece of photographer's tintype into strips about 1 inch wide and 3 inches long, then cut away all except enough for a handle, leaving 1 inch square on one end; bend the end of this handle, and fasten into a cork in a bottle, which will serve for a holder. Upon this plate place the clean cover,

and by means of a pipette drop a little of the dilute alcohol and diatoms upon it, applying a gentle heat with a spirit-lamp. The alcohol takes fire and burns off. The remaining alcohol causes the diatoms to become evenly distributed. If inclined to mat, touch with a hot pin or needle. Now bring the whole to a red heat for plenty of time to make the diatoms appear white and perfectly clean.

Now take a thin glass slip upon which a very thin cell of brown varnish has been made and well aged as previously described, take up the thin cover with diatoms attached and place it upon the slightly gummed end of a cedar-wood pencil, the diatomed side being uppermost. Now with a fine sable pencil coat the edge of the thin cover with a layer of thick brown varnish, wait until it has nearly set, and invert it carefully and firmly upon the cell-walls of the slip. A slight pressure with the pencil will cause the cover to adhere all round, when the slide may be set aside for about half an hour ; it must then be placed on the turntable and a slight coat of brown varnish applied to the edge of the circle.

The slide should now be put away for a week or more, after which time it should be examined with the power suitable to it and if not satisfactory, discarded. If, on the other hand, it is a good preparation, the student may proceed to finish it by placing it on the turntable and describing a ring of white zinc varnish so as to cover the edges of the circle, making it completely air-tight. When this is dry, various coloured rings may be turned upon the white substratum, giving the whole a very pleasing appearance.

Some writers have deprecated this ornamentation of slides, but the author's opinion is that the time is well spent over a good slide, while a bad one, or even one of medium quality, should be washed off as soon as examined for the first time. The composition of the coloured varnishes may be found in the list of recipes at the end of this chapter.

The process of arranging diatoms is a simple one, easily done by means of a small camel's-hair pencil, with the aid of the erector and a half-inch objective. The brush should be drawn through the lips, and when using the microscope-stand shown in Fig. 24, *sitting well over it*, the side of the hand only resting on a support, it is quite easy after a little practice to remove any diatom from the field of view. For this purpose many workers use what is called a mechanical finger as a substitute for the fingers of the human hand. Zentmayer's form has been described in the 'Monthly Microscopical Journal,' and is here reproduced as Fig. 239.

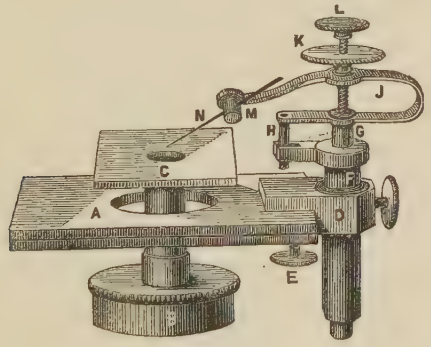


FIG. 239.

Rezner's form of this instrument (Fig. 240) is much more simple than the above, and may be adapted to any microscope. It consists, as may be seen in the engraving, of a sleeve, which is passed up over the objective just far enough to possess a firm bearing, and so that the point of the bristle is in focus when depressed to nearly its full extent. In order to use this finger, the point of the bristle should be brought into the centre of the field, touching the object slide, and then withdrawn, still in the axis of the microscope, until it has become invisible. The desired object is

then sought for, such as diatoms, foraminifera, or other minute specimens, and brought into the centre of the field; the point of the bristle is then lowered by the screw until it touches the desired object, which usually adheres to it at once.

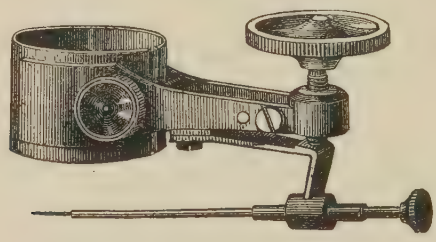


FIG. 240.

Some operators recommend a cat's whisker for the purpose of using with the mechanical finger, and no doubt it is as good as anything. Still the worker with this piece of apparatus will find that often, especially when working with ordinary bristles, it is almost impossible to deposit the object exactly where it is required, so pertinaciously does it adhere to the bristle. Prof. H. L. Smith has advised the use of a slender thread of glass for this purpose, but taking all things into consideration the cat's whisker seems to be the best.

The diatoms are picked one by one and placed where desired. To prepare the slide, put it on a turntable, and with a pen make a small circle in the centre to guide in placing. On the reverse of the slide put a tiny drop of pure distilled water, with a small fragment of gelatine in it; so that when the diatoms are arranged, breathing on them will bind them into the size. Foraminifera, Polycistina, and other similar objects may be arranged in the same manner.

As another instance of dry preparation may be given a method for mounting starch granules. The starch, say

from the potato, should be well washed from all foreign substances, and mixed with cold distilled water, so as to form a slightly opalescent liquid. A thin glass cover having been cleaned is breathed upon, laid level upon the table of the desiccator (Fig. 238), and a drop of the starch solution deposited upon it. When perfectly dry it may be coated at the edges with brown varnish, and inverted upon a shallow varnish cell, as has already been described for diatoms, and finished in the same manner.

Such specimens as micro-fungi, *Penicillium glaucum* (*l*), *P. roseum* (*n*), *Ascophora mucedo* (*j*), *P. chartarum* (*k*), *Aspergillus glaucus* (*m*), Fig. 241, can only be successfully

FIG. 241.

mounted by the dry method, as when mounted in fluid the spores become detached, and the chief characteristics are lost.

The best way to mount such specimens would be to choose a cell deep enough to hold them comfortably, and

place a small drop of Farrant's medium in the centre, to which is attached a portion of the substratum bearing the fungus upon it. The slide must now be placed under the desiccator until perfectly dry, when the thin covering glass should be placed on in the manner already described, and finished in the usual way.

There is another style of mounting it may be useful to describe, i. e. in such a manner that the Lieberkuhn may be used with the slide, to produce illumination on a dark ground, so that such objects as Foraminifera or the pollen of the mallow may be viewed with this appliance, as an opaque object.

Take a very shallow varnish cell slip, but deep enough of course to contain the object, and turn upon the centre of it a disc of Bate's dead-black varnish, allow this to dry and become well aged. When proceeding to mount, say the pollen of *Althea rosæ* (Fig. 97), dust it over this blackened disc, place in the desiccator until dry, and then put on the cover in the usual manner. If the pollen be dusted over the whole of the circle, the observer will be able to use the slide both as a transparent and opaque object. Many preparations, such as the disc of deal, do not require fixing either to the cell or its cover, and this class of objects requires no special directions for successful mounting.

MOUNTING IN GUM RESINS.—Objects required for mounting in gum resins need of course quite as much preparation as when put up by the dry method; but that we are about to describe is perhaps the best suited for beginners, as the gum resins are good preservatives, and the preparations are likely to be permanent with the minimum amount of care.

Canada-balsam has been used for a long period for this style of mounting, either by itself or when diluted with chloroform or benzol.

We will here go through the operation of mounting a rock section in undiluted balsam, just to illustrate the only case in which the author advises the use of papers to cover up the whole of the slide.

A hot-water plate is used by a great many mounters. It may be a tin box 9 inches square by 2 inches in depth, supported on a stand or upon four legs. In one corner is a funnel-shaped neck for the introduction of water and the escape of steam when in use, the chest being heated by means of a gas-burner or spirit-lamp.

Bell-jars are sure to be required for many purposes, but principally for keeping the dust from objects which are undergoing preparation. They need not be large, and broken wine glasses may be well utilised for this purpose if the stem only is broken.

When speaking of the hot-water box, perhaps we should have mentioned the hot plate, which is still a favourite with many mounters, who still cling to undiluted balsam. This is a brass plate, standing upon four legs, and is usually heated by means of a spirit-lamp, though there is no reason why it should not be heated by gas.

The reader will remember that when treating of section-cutting, the surface of the slice first polished was to be cemented firmly to a glass slip, the final grinding being performed upon it. When grinding a section to extreme thinness, the slip is often badly scratched, and thus disfigured; some operators advise the section to be taken off this slip, and put upon another for permanent mounting, but in many cases this is not admissible, and the following method should be adopted. Take the glass slip with the section upon it and clean the surface with water, and after with methylated spirit; place a tiny drop of balsam (rendered fluid by heat) upon the centre of the section, carefully put on the cover and place the whole on the

hot plate, gently press down the cover, keep on the hot plate for a short time and take away to a cool place to set.

No more balsam should be used than sufficient to reach to the edges of the glass cover, and if this point be carefully attended to, the slip will require no cleaning preparatory to covering with paper.

MOUNTED BY
GEORGE E. DAVIS

Boiler Incrustation.
(*Section.*)

Catalogue No. 611.
Date 10/3/80.
Medium—
Balsam.
Cell .....

The covers used and recommended by the author may be seen as annexed.

These covers should be cut about $\frac{1}{32}$ of an inch less than the slide they are intended to cover, leaving the edges of the glass exposed; there is no use in binding up the glass in paper, it only helps to absorb moisture, and in the case of many objects mounted dry, serves to encourage the growth of mildew.

When mounting in pure balsam, especially if old, the slide gets very hot, so that the American clip shown in Fig. 242 is a convenient article.

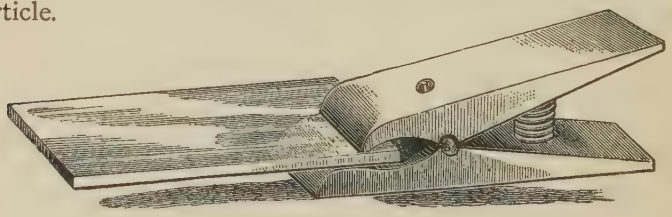


FIG. 242.

It has already been shown under the head of dry mounting how diatoms are distributed over the surface of a thin cover. They may also be mounted in balsam.

On the centre of the glass slip place a tiny drop of old balsam, and with a pair of tweezers place the cover-glass

over it and hold the whole over the spirit-lamp until a sea of the bubbles is seen underneath. Remove, and with a gentle pressure press down the cover. The bubbles will all disappear and the balsam become hard. To secure the diatoms all in the same plane, turn the cover-side downwards, and leave in a warm place. This is best effected in the whalebone clip shown by Fig. 243, but care should be taken that the balsam does not project beyond the

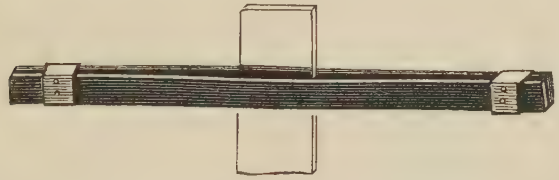


FIG. 243.

cover, or it will stick to the whalebone, and cause considerable annoyance.

The old plan of mounting objects in pure Canada balsam has been almost abandoned; it is now usual to put up objects in balsam and benzol whenever balsam-mounted objects are required.

To illustrate how this should be done, let us proceed to mount one of the forelegs of the great water-beetle (*Dytiscus marginalis*) shown in Fig. 244.

After detaching the leg of the insect, the first operation is to soak it in potash solution for a day or two, then take it out and wash it in water, allow it to soak in dilute spirit (1 of spirit to 3 of water) for 24 hours, and then transfer it to methylated spirit. After remaining here until all the moisture has been extracted by the spirit it must be taken out, drained on blotting paper, and placed in oil of turpentine. Here it must remain until the colour of the chitinous skeleton has become sufficiently reduced, when it

may be soaked for a few hours in benzol to remove the excess of turpentine. It is now ready for mounting. Place a very small drop of balsam and benzol upon the centre of a glass slip, take up the leg by means of a pair of forceps, and place it upon the drop of balsam; now put another small drop of the medium upon the object, and taking up the well-cleaned covering glass with the forceps drop it carefully upon the object in a perfectly horizontal manner. Now *gently* press down the cover with a camel's-hair pencil in such a manner that the object is not disturbed, and put on the spring mounting clip, as shown in Fig. 245. The slide should now be heated over a spirit-lamp until the benzol *just commences* to boil, when it must be removed to a cool spot to thoroughly set.

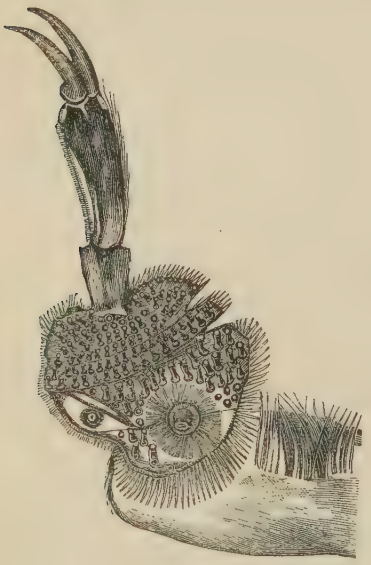


FIG. 244.

FIG. 245.

The author's practice is, now, to set the slide on one side for a few weeks, then to clean off the excess of balsam with a scalpel, finally cleansing by slight friction with a piece of cotton wool or sponge moistened with methylated spirit. After another repose for a few days, the slide is placed on the turn-table, and a coating of brown varnish applied, so that the circle embraces the edge of the cover and the slide also,

when it may be finished with the white zinc varnish and coloured rings as already described.

Balsam and benzol may be used cold, but as the operation is exactly similar to that of mounting in dammar and benzol it will not be further described.

The operation of mounting wheat-starch in dammar and benzol will sufficiently explain the cold process of mounting objects in gum resins. Make a mixture of wheat-starch and distilled water, as before advised for potato-starch; place a drop of this upon the centre of a glass slip, and put in the desiccator (Fig. 238) to thoroughly dry. Now take a drop of benzol and place upon the starch, and before it has time to thoroughly evaporate drop on a little dammar and benzol, put on the glass cover, place the spring clip upon it and set aside for several weeks, when the excess of dammar may be removed and the slide finished as already described. Where many objects are required to be mounted at once, spring clip boards may be used. Many forms have been devised, but that figured in 'Science-Gossip' for October

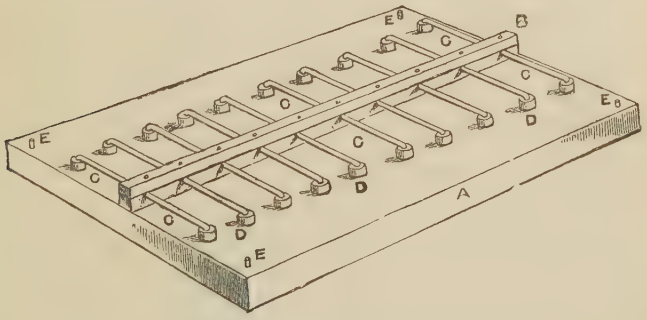


FIG. 246.

1881 is one to be preferred. It is shown in Fig. 246: A being a base-board of mahogany, C pieces of clock-spring bent at each extremity and fitted into corks at D. At E are four screws which admit of several of these boards being

placed above each other without coming in contact with the springs.

This board may be found of much use, but it is certainly better in many instances to have small independent clips capable of being placed in the hot chamber, air-pump, and similar situations with the slide itself. Many objects may be mounted in gum resins, and the dammar and benzol will perhaps be the best substance *generally* for objects requiring to be shown with the aid of the polariscope.

Sometimes with gum resins a source of annoyance is the appearance after a time of a white cloudiness, completely ruining many otherwise carefully mounted specimens. This is caused by dampness, or the presence of fatty matters not carefully removed before applying the balsam. If the foregoing instructions be carefully followed this mishap can scarcely happen.

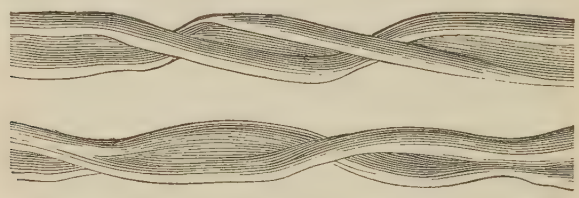


FIG. 247.

Cotton fibres and sections of the same, shown in Figs. 247 and 248, make good polariscope objects when mounted in

FIG. 248.

dammar and benzol; they should be soaked in spirit to remove water, transferred to benzol and mounted direct from that fluid.

Dust and dirt are the greatest enemies of the microscopist, and every operation which can be done under cover

should be so arranged. In these cases very small but wide-mouthed bottles are conveniences, watch-glasses, small beakers, and stemless or footless wine-glasses will always come in useful during these operations; it should be the aim of the student to be cleanly in his manipulations, and endeavour to procure the best results with the minimum expenditure of material.

Mounting insects without pressure has been much practised of late, and the splendid preparations of Mr. F. Enock cannot be passed by unnoticed. A method of doing this may be found as follows:—Let us take the head of the *Dytiscus marginalis*. Soak it for two days in equal parts of spirits of wine and water, after which transfer it to absolute alcohol for two or three days longer; now transfer it to turpentine and place it in the light until fairly bleached, take it out of the turpentine and place in benzol until all the former has been eliminated. Now choose a cell just deep enough to hold it, glass or pure tin preferred (by no means must it be of brass, this metal being acted upon by balsam), rinse it out quickly with benzol, insert the head and pour in some balsam just rendered thin enough to run, with benzol, until the cell is full, now put on the cover, cleanse the edge of the cover with a brush dipped in benzol or spirits of wine, and set aside in a warm place that the benzol may escape and become hardened round the edge of the cover. When fairly dry give a good coat of guaiacum varnish (see Recipes), and when this is dry repeat the coating. Finally finish the slide with a ring of asphalte, or black guaiacum varnish.

MOUNTING IN AQUEOUS MEDIA.—This style of mounting comprises the fluids and semi-fluids or viscid media; perhaps the latter are the easier done, but for sake of order an illustration of how to mount starches in carbolised water (see Recipes) will teach the method.

A suitable cell must first be selected, one of brown varnish in this instance, and filled with the carbolised water just made milky with the starch. The thin glass cover having been taken up on the end of a pencil as already described when mounting diatoms by the dry method, is edged with brown varnish, allowed a few minutes to nearly set, and then placed in contact with the cell but not pressed down in the centre to any extent. The superfluous fluid may then be absorbed with blotting paper, the slide placed on the turntable, and another coat of brown varnish applied to the edges. Put aside now for a day or more, and then finish with white zinc varnish or asphaltum according to the taste of the operator.

If the student possesses an air-pump he will find that fluid mounts will be rendered more permanent by placing the cell filled with fluid under the receiver so as to eliminate the air which most liquids contain. The best form of air-pump has been shown at Fig. 222, but there are other forms which, though not so generally useful, are nevertheless handy on account of their small size. One of these is shown in Fig. 249, from which it will be seen that it is capable of taking slides only.

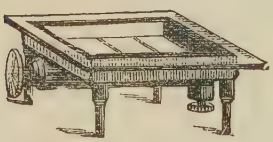


FIG. 249.

Many other fluids may be used in place of the carbolised water: the cuticle of esparto grass is shown very well when mounted in dilute acetic acid, the fibres of jute exhibit the ladder-like markings to perfection when put up in dilute spirit, while many of the desmids and minute algæ can only be kept in distilled water in which a lump of camphor is kept. Some of the smaller organisms may be put up in a very dilute solution of osmic acid. This plan is advised by Mr. Saville Kent for the preservation of the collared monads, in his 'Manual of the

Infusoria,' such as the *Monosiga Steinii* and *Salpingœca convallaria*, shown in Fig. 250. He states that they may be sealed up after treatment with osmic acid without the addi-

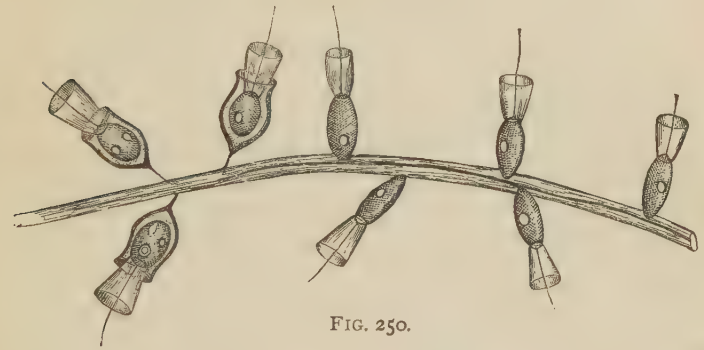


FIG. 250.

tion of any other preservative, and that it will be found the smaller and most delicate flagelliferous species are equally amenable to this treatment, preserving their flagella, and even, in the case of the Choano-Flagellata, their sarcode collars in a life-like form. Many fluids have been described for these purposes, some will be given in the recipes at the end of the book, but the beginner should remember never to use a fluid in a cell unless he is thoroughly acquainted with the action the fluid has, or has not, upon its walls or on the cements or varnishes used in connection with it. If these points were more studied, fluid mounts would not be looked upon with so much ill-favour as they are now.

Objects mounted in glycerine should *always* be put in a cell, and if the cover be put on in the manner described for mounting in carbolised water, and diatoms, dry, the slip can be easily cleaned from any superfluous glycerine. After this, put the slide on the turntable, and describe a neat ring of brown varnish, finally finishing with a *broad* ring of white zinc varnish, as before described.

Objects for mounting in glycerine, such as a portion of muscle with Trichinæ *in situ* (Fig. 251), should be soaked for some time in a mixture of equal parts of water, alcohol, and glycerine, then exposed to the air under a bell-jar, or in the desiccator; the tissues gradually become filled with strong glycerine, and the object is then ready for mounting in that medium.

FIG. 251.

Turning now to viscid media, the author has made much use of Farrant's medium, having mounted mosses and starches very successfully in it. It is a very convenient substance, seeing that it is used cold, and the slide cleaned with water and a camel's-hair brush *immediately* after mounting; when on the following day the cover may be finished with white zinc varnish and the usual coloured rings.

The next description of mounting, that in glycerine jelly, is one which, to a beginner, is frequently a stumbling-block. It is an exceedingly convenient method to adopt when the object would be rendered too transparent in balsam or dammar, or when it is undesirable to dry it at all.

In the preparation of the many things from the vegetable kingdom, as mosses, algæ, cuticles, and sections, and from the animal kingdom, as many eyes and wings of insects, gastric teeth, palates of the Mollusca, it is only necessary, if they are sufficiently clean and not too dark in colour, to put them for a few hours into a mixture of methylated spirit, glycerine, and water (about equal parts of each), although exactness is not necessary, as the mixture may be varied to suit circumstances.

When they are taken from this mixture, they must be placed upon the centre of the slide, and the surplus fluid

absorbed by blotting-paper. Either of the two plans may now be followed with regard to the jelly—it may be liquefied by placing the bottle in hot water, and then dropping the liquid jelly upon the slide, or a small piece may be cut from the bottle and put upon the object and the slide gently warmed, when the jelly will diffuse itself through the object, and will be found exceptionally free from air-bubbles; but should there be air-bubbles or not, it is of great value to boil the jelly and object upon the slide, but care must be used or the mount may be ruined. Should the boiling be decided upon, the clip should be used, and the slide held over the flame of a lamp; it will at first begin to bubble from the centre outwards, and if the slide be carefully watched, a very perceptible crack may be seen and heard: at this moment, and without delay, the slide must be withdrawn from the heat and placed upon a cold surface (an iron slab for instance), when the jelly will rapidly set and air-bubbles be excluded.

The mounts are easily cleaned from superfluous jelly, by brushing with a *soft* tooth-brush under a running water tap, the surface of the slide being allowed to dry spontaneously. It will be found that the slide is free from glycerine smears, which interfere much with the after-process of finishing.

FIG. 252.

The slides may then be finished by ringing with white zinc varnish and the various coloured circles.

Such is the method employed for mounting in glycerine

jelly, and very satisfactorily illustrates the general practice, though of course it is not all objects which will bear boiling.

An instrument called Smith's mounting machine, and shown in Fig. 252, is very useful when mounting large and elastic substances in glycerine jelly, as by it any degree of pressure may be exerted upon the covering glass.

The method of using this instrument may be explained by mounting a lichen section (Figs. 253 or 254) in Deane's medium. The section having been cut as thinly as possible, it is to be soaked for a day or more in some of the medium diluted with just enough water to render it fluid; it is then to be placed upon a slip, the cover superposed and

FIG. 253.

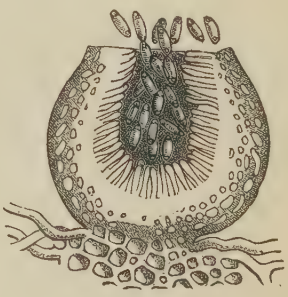


FIG. 254.

placed in the mounting instrument, a gentle pressure being exerted by means of the screw; the slide must now be warmed over the lamp, and a drop of the medium placed at the edge of the cover, when it will be drawn under by capillary attraction. This is a very clean

way of mounting specimens, and one often used by the author.

The foregoing instructions have been given more as an aid to the student than to the expert mounter. It would be impossible to give processes for mounting everything, as even the preparation varies in nearly every specimen. All that has been said to enable the beginner to practise the art of mounting has been written in a general sense, and each process must be carried out *with intelligence.* The student should endeavour to make his preparations look as natural as possible, and no pains should be spared in this respect. Again, the question of varnishes and cements is one which demands careful study, and the beginner is advised to well peruse Chapter XIV. before he commences actual work.

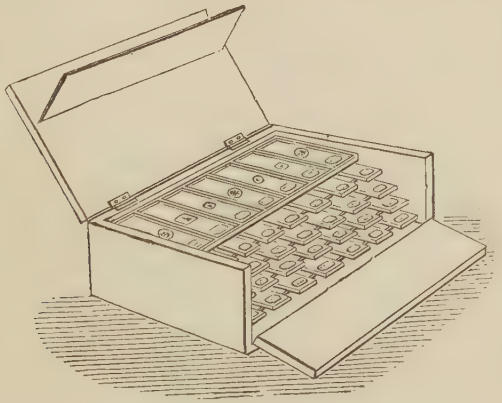


FIG. 255.

When the slides are mounted, a cabinet is required to keep them in. They are generally made to one pattern, and may be obtained from any dealer in microscopical apparatus. A special and cheap form of slide box was devised some years ago by Mr. Stokes. It is shown in Fig. 255, and it

may be observed that the labels upon the slides are readily seen without the trouble of taking out each tray.

In special methods of microscopical research there will be wanting many fine and delicate pieces of apparatus, which ofttimes will have to be made and devised by the student himself. This is excellent practice, and he should learn to exercise his ingenuity to the utmost, and not be running to the optician constantly for every trifling little thing he requires. An instance of how apparatus may be improvised is shown by Fig. 256, which is a breeding cage for Podura,

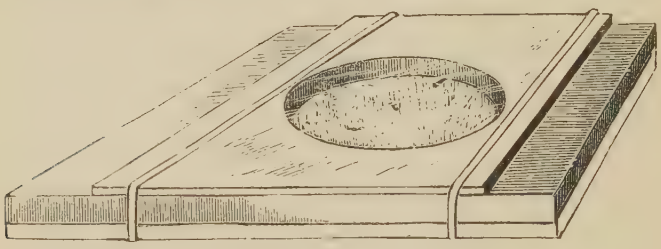


Fig. 256.

devised and illustrated in the 'Monthly Microscopical Journal' by Mr. McIntire. The glass plate, perforated with a hole, is covered on each side with a thinner glass, and held in position by two indiarubber bands. This is very useful when the scales of these insects are required, and it may also be brought into use as a flea cage, and for many other purposes.

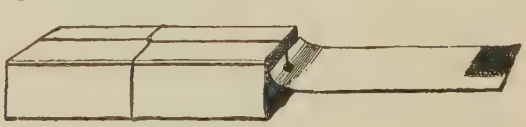


Fig. 257.

In concluding this chapter a few words may be said upon sending slides by post. Pack them in a small wooden box

to be obtained of any optician, tie it securely with fine string and pack in a *black* paper wrapper. Attach a luggage label to one end, upon which is to be written the full address, and upon it place the postage stamps. No writing of any kind should be put upon the box itself in order that it may reach its destination in safety. The whole when ready for the post should appear as in Fig. 257.

CHAPTER XIV.

REAGENTS—RECIPES.

LITTLE now remains but to notice the action and properties of some of the various reagents used for microscopical purposes, and in order to put them in something like order, they are divided into four classes—softening agents, dehydrating and hardening agents, bleaching and oxidising agents, and solvents.

Softening Agents.—One of the mildest softening agents with which the microscopist has to deal is acetic acid diluted with four times its weight of water ; it renders some tissues quite transparent, and used in conjunction with glycerine serves for sending insects for dissection to long distances. Acetic acid dissolves phosphate and carbonate of lime, but does not dissolve oxalate of lime, and neither has it any action in the diluted state upon any of the varnishes or cements in the general use of the microscopist.

Glycerine is a fluid of the greatest use in microscopical research, particularly as an agent for preventing the drying up of tissues. One part of pure glycerine diluted with nine parts of water produces a fluid of the same density as sea-water, and this strength is a very handy one to use. Contrary to the general opinion, cells filled with glycerine do *not* necessarily leak, it is only when objects have been *slovenly* mounted in this medium that such a mishap occurs.

The alkalies, potash, soda, and ammonia, are somewhat alike in their action upon many substances, ammonia being

the weakest reagent. When used in concentrated solution, animal substances, especially of an albuminoid nature, are entirely dissolved, while when used in a weaker form many organs are separated in such a way as to be readily obtained. Upon vegetable substances the alkalies act somewhat similarly, the leaf the esparto grass (*Macrochloa tenacissima*) and the straw of cereals, and even blocks of pine wood, are all softened and disintegrated by boiling in a solution of caustic soda containing 50 grains to the ounce of water. Dilute nitric acid (one part of acid to ten of water) may also be used as a softening agent in certain cases, and, it may be added, should be used in glass or porcelain vessels, as metals are rapidly dissolved by this acid. This remark applies also, though in a lesser degree, to acetic acid.

Dehydrating and Hardening Agents.—Dehydrating agents for use with the desiccator (Fig. 238) are not put in contact with the preparation; but placed below in a separate compartment underneath a bell-jar, as shown in the figure. Anhydrous phosphoric acid is perhaps the most powerful absorbent we have, and next to this in usefulness is concentrated oil of vitriol.

Fluids, however, are not so handy as solids, and therefore for general work *fused* chloride of calcium, quick-lime, or carbonate of potash, are more generally used.

When, however, a substance requires to be dried or dehydrated without losing its fresh or moist appearance, it must be brought in contact with a water-absorber, such as alcohol. Absolute alcohol is an excellent fluid for this purpose, and is not equalled by any other reagent; methylated spirit is often used in its place, and for many purposes will suit admirably.

It must not be forgotten that alcohol has a more or less solvent action upon most of the gums used for micro-

scopical varnish making, and therefore strong alcohol cannot be used in such cells. Weak spirit has, however, no action upon a cell of gold size, and this is the only varnish the author advises to be used when an object is mounted in weak spirit.

As to hardening agents, the chromates have been treated of in Chapter VIII. on Section-cutting, and therefore it is not necessary to recapitulate their uses.

Tannin dissolved either in water or alcohol is a hardening agent especially useful for gelatinous tissues; it is used sometimes in injecting for hardening the walls of blood-vessels to prevent the passage of the coloured gelatine through them. Alcohol itself is a very powerful but safe hardening agent, and when combined with tannin may find many uses.

Bichloride of mercury solution has been recommended as a hardening agent, and it probably acts by forming insoluble compounds with the albuminoid matters. It is so deadly a poison that the author can scarcely advise the student to use it.

Osmic acid, easily purchased in a one per cent. solution, is also a splendid hardening agent, and is of great use in studying the lower forms with naked protoplasm. It is, however, very poisonous. By the action of this reagent the currents in the protoplasm of Myxomycetes are instantly suspended, and in a few moments the plasmodium is sufficiently hardened to make sections possible.

Bleaching and Oxidising Agents.—Under this class we have :—

Chlorinated soda (eau Labarraque),
Chloride of lime (bleaching powder),
Chlorate of potash,
Bichromate of potash,
Nitric acid,
Turpentine,

all of which have their special uses.

The first two are principally used for bleaching vegetable sections, and the strength of the solution may be found in the Receipts at the end of the book. It is most important, when using these reagents, to eliminate every trace of them after the operation is finished, and this may be done by soaking in a bath of neutral sulphite of soda, 15 grains to the ounce of water; a solution of one part of strong liquid ammonia to twenty parts of water will also effect this.

Chlorate of potash is generally used for cleaning diatoms, in connection with some strong acid such as sulphuric, nitric, or hydrochloric. Great care should be used in making these mixtures, as it is easy with them to produce explosions, and their use should (in connection with chlorate of potash) be avoided as much as possible. Nitric acid by itself is a powerful bleacher and oxidiser, but has the disadvantage of emitting powerfully noxious and acid fumes; when slightly diluted to prevent this, its oxidising powers can be increased by the addition of chromic acid or bichromate of potash. This admixture is not in the least likely to explode.

Another extremely powerful oxidiser is a mixture of strong sulphuric acid and chromic acid in crystals; it is especially useful in cleansing diatoms, and must be heated carefully over the spirit-lamp to obtain its maximum effect. Peroxide of hydrogen is an extremely valuable bleaching agent and oxidiser, and likely to come into general use when its properties are better known to microscopists; it is neither acid nor alkaline, and does not give off any objectionable odour. Another bleaching agent is turpentine, almost exclusively used to reduce the intensity of colour in the chitinous skeletons of insects. After having been treated with potash or soda solution, and the abdominal contents expressed, the insect is washed with water, dried in alcohol, and allowed to soak in turpentine until the colour of the chitin is sufficiently reduced in intensity. Turpentine has

a solvent action upon many of the varnishes used for microscopical purposes, and therefore great caution should be exercised in this respect.

Solvents.—Under this heading come many reagents of diverse character: we have alkalis, acids, and neutral compounds, all of which have their special applications. Amongst the first class are solutions of potash, soda, and ammonia, having a great affinity for most animal substances. Grease unites with any of these, producing a soap which easily dissolves in water. The alkalis in their solid state should be handled with caution, owing to their corrosive nature; they are chiefly used in solution to dissolve out the internal organs of insects, and to prepare the leaves of mosses for exhaustive scientific examination.

The acids, sulphuric, nitric, hydrochloric, and acetic, have each their uses; for dissolving metals, oxides, carbonates, phosphates, and other salts, they are necessary. They have no action upon silica, but upon animal and vegetable fibres the action is very decided.

By far the most important solvents we have to consider are ether, alcohol, benzol, glycerine, oil of cloves, oil of cajeput, and last, and not least, water. Ether is not very soluble in water, it dissolves in about fourteen parts at the ordinary temperature of the air; it carries into solution many organic compounds, as the volatile oils, resins, fats, alcohols, tannic acid, which are but sparingly soluble in alcohol, while it is without action upon many substances easily dissolved by that reagent. It is miscible in all proportions with alcohol, bisulphide of carbon, and naphtha.

Alcohol is probably the most used fluid in microscopy; it is miscible in all proportions with wood-spirit, chloroform, acetic acid, and naphtha, and is a good solvent for most resinous substances. Attention should be paid to the strength of the alcohol; alcohol absolute is expensive,

and though a good drier, sometimes fails in its action because it is too strong. Ordinary druggists' alcohol of 85 per cent. is an excellent solvent for resins, camphor, tannin, the balsams, iodine, acetic acid, and castor oil, but the ordinary "spirits of wine," consisting of equal volumes of druggists' alcohol and water, has not the same solvent action for resinous matters. Methylated spirit consists of ordinary alcohol, mixed with 10 per cent. of wood-spirit, and is capable of being put to the use of ordinary alcohol by the microscopist.

Benzol is a colourless, strongly refracting liquid, of a very inflammable nature. It is almost insoluble in water, but dissolves freely in alcohol and ether. It dissolves iodine, sulphur, fats, gum-resins, and many other compounds, such as caoutchouc and guttapercha.

Petroleum naphtha, called also benzoline, benzine, gasoline, &c., is nearly as useful as coal-tar benzol for the purposes of the microscopist, provided it be of good quality and does not contain any quantity of heavy oils.

Glycerine is a solvent strongly recommended for general use by Dr. Beale. It is soluble in all proportions in water and alcohol, though it is but sparingly soluble in ether. It dissolves nearly all organic substances soluble in water, and many of those soluble in alcohol; it dissolves small quantities of carbonate of lime and many oxides; 20 per cent. of arsenious acid, and 10 per cent. of tannic acid, forming with this latter a waxy solid, melting at the temperature of the body, which the author has used sometimes for mounting purposes.

Oil of cloves is often used as an intermediate bath between alcohol and Canada balsam; it serves to take out the alcohol, with which the oil changes place. One volume of oil of cloves dissolves an equal volume of alcohol.

Oil of cajeput is often recommended for use in place of

oil of cloves, and is in every way cheaper, as one volume of it absorbs eleven volumes of alcohol, or is eleven times more efficient than oil of cloves.

And now we must consider water, perhaps the most useful solvent we have. A great many substances are soluble in this fluid, and even those generally considered insoluble are acted upon to such an extent as to interfere with it for microscopical purposes. Air, or rather the oxygen and nitrogen of the air, are soluble in water, and cause the microscopist no little trouble. This has been dealt with on page 304, and it may also be stated that when water is to be evaporated on a slide for future mounting, the water should be distilled, so that no objectionable residue is left.

RECIPES.

Carbolic Acid (*Fluid*).

Procure an ounce bottle of Calvert's pure crystallised carbolic acid, place it in a jar of warm water to melt; then add two drachms of methylated spirit, mix, and preserve for use.

If strong carbolic acid be spilled upon the hands, it must be *immediately* wiped off with *oil*, not water.

Carbolic Acid Water.

Strong.

Fluid carbolic acid	1 drachm.
Distilled water	16 oz.

Weak.

Carbolic acid water (strong)	1 oz.
Distilled water	9 oz.

Chlorinated Soda (*solution*).

Dry chloride of lime	2 oz.
Soda crystals (washing soda)	3 oz.
Water	2 pints.

Mix the chloride of lime with half the water, and the soda in the other half, mix the whole together, and allow to settle in a well-corked bottle. Pour off the clear liquid for use, which must be kept in a well-corked bottle.

Chlorinated Lime (*solution*).

Dry chloride of lime	½ oz.
Water	1 quart.

Dissolve and allow to settle.

Liquor Potassæ (*solution*).

Caustic potash (*in sticks*)	2 oz.
Water	1 quart.

Caustic soda may be substituted for the potash.

Iodine Solution.

Iodine	40 grains.
Iodide of potassium	60 grains.
Water	1 pint.

Dissolve the two substances in four ounces of the water, then add the remainder.

Gum Water.

Gum arabic	4 oz.
Glycerine	¼ oz.
Weak carbolic acid water	4 oz.

Allow to stand in the cold until dissolved.

This will be found an excellent medium for attaching labels to glass.

Spicer's Fluid.

Alcohol	3 fluid oz.
Distilled water	2 fluid oz.
Glycerine	1 fluid oz.

Goadby's Fluid, No. 1.

Bay salt	4 oz.
Alum	2 oz.
Corrosive sublimate	4 grains.
Water	2 quarts.

Dissolve and filter, when it will be ready for use.

Goadby's Fluid, No. 2.

Bay salt	8 oz.
Corrosive sublimate	2 grains.
Water	1 quart.

Camphor Water.

Distilled water	1 quart.
Tincture of camphor	1 drachm.

Well mix, but use only the clear fluid.

Thwaites' Fluid.

Rectified spirit	1 oz.
Creosote (wood)	q. s.
Distilled water	1 pint.
Precipitated chalk	q. s.

Saturate the spirit with creosote, then add the water and a little precipitated chalk. Filter, when the liquid is ready for use.

Ralf's Liquid.

Bay salt	1 grain.
Alum	1 grain.
Distilled water	1 oz.

Glycerine and Acetic Acid.

Glycerine	2 oz.
Glacial acetic acid	½ oz.

Glycerine and Gum.

(*Farrant's Medium.*)

Gum arabic	4 oz.
Distilled water	4 oz.
Glycerine	2 oz.

Dissolve in the cold.

Deane's Medium.

Nelson's gelatine	1 oz.
Honey	5 oz.
Creosote	6 drops.
Alcohol	½ oz.
Water	5 oz.

Soak the gelatine in 4 oz. of the water for twelve hours, add the honey, previously heated to nearly boiling point, in a separate vessel, and boil the whole together. When cooled somewhat, add the spirit and water in which the creosote has been dissolved. Afterwards, filter through fine flannel.

Glycerine Jelly (*Lawrance's*).

Gelatine	1 oz.
Glycerine	6 drachms.
Camphorated spirit of wine	¼ oz.

Cover the gelatine with cold water, and allow it to soak until it becomes soft. Dissolve by placing the jar containing it in a vessel of boiling water, and allow it to cool, then add a small quantity of the white of an egg, and boil the mixture until the albumen coagulates, when the whole is to be filtered through fine flannel, and mixed with the glycerine and spirit.

Liquid Glue.

Dissolve shellac in wood-naphtha at a very low temperature, until the mixture is of the required consistency.

It makes a very brittle varnish, but very good cells.

Brown Varnish.

Pure indiarubber	20 grains.
Bisulphide of carbon	q. s.
Shellac	2 oz.
Methylated spirit	8 oz.

Dissolve the indiarubber in the smallest possible quantity of bisulphide of carbon, and add this to the alcohol in such a manner that the whole is mixed without the formation of clots. Now add the shellac, and place the jar containing the mixture in boiling water, until the whole of the shellac is dissolved, and the smell of the bisulphide has disappeared.

Guaiacum Varnish.

Gum guaiacum	2 oz.
Shellac	2 oz.
Methylated spirit	10 oz.

Powder the guaiacum and dissolve it in the spirit, filter, and then add the shellac. Keep the whole in a jar surrounded by warm water until dissolved.

Matt Varnish.

Gum mastic	40 grains.
Gum sandarac	160 grains.
Methylated ether	4 oz.
Benzol	1½ oz.

Black Matt Varnish.

Gum mastic	50 grains.
Gum sandarac	200 grains.
Methylated ether	1½ oz.
Benzol	½ oz.

Dissolve the gums in the fluids and triturate in a mortar with sufficient lampblack of the finest quality.

Marine Glue.

Indiarubber shreds	2 oz.
Shellac	2 oz.

Dissolve the indiarubber in solvent mineral naphtha, add the shellac in powder, and heat until liquefied, well mixing the whole together. It produces a solid marine glue, and requires heat in its application. The author has it on good authority that very little of the marine glue at present made contains a particle of indiarubber.

Photographic Varnish (*for negatives*).

Gum benzoin	½ oz.
Gum sandarac	1 oz.
Methylated spirit	12 oz.

French Polish.

Shellac	3 oz.
Gum sandarac	½ oz.
Methylated spirit	1 pint.

French polish is sometimes coloured with gum dragon, &c.

Brunswick Black or Black Japan.

Egyptian asphaltum	4 oz.
Linseed oil	4 oz.

Boil together for some time, and mix to the required consistency with oil of turpentine.

Canada Balsam Varnish

(*For rendering ground glass transparent*).

Take 4 oz. of Canada balsam, and bake in a cool oven till quite brittle when cooled. Dissolve this in 12 oz. of benzol, in which ½ oz. of mastic has been previously dissolved.

Black Varnish (*Davies*).

Indiarubber shreds	30 grains.
Egyptian asphaltum	4 oz.
Solvent naphtha (mineral)	10 oz.

Dissolve the indiarubber in the naphtha, add the asphaltum, using heat if necessary.

Balsam and Benzol.

Bake the Canada balsam in a cool oven, and then dissolve to the right consistency with benzol. If baked too long the residue will be too brittle ; if too short, it will not dry quickly enough.

Dammar and Benzol.

Gum dammar	1 oz.
Benzol	2 oz.

Mastic and Benzol.

Gum mastic	4 oz.
Benzol	3 oz.

Gold Size.

Linseed oil	25 oz.
Red-lead	1 oz.
Powdered white-lead	q. s.
Yellow ochre	

Boil the oil and red-lead together for about three hours, taking care it does not burn or boil over; pour off the clear fluid, and boil again with a mixture of equal parts of the white-lead and yellow ochre, added in small successive portions. Pour off the clear fluid for use.

Black Gold Size.

Triturate in a mortar 1 fluid oz. of gold size with sufficient lampblack to form a dense black colour. If too thick, it may be thinned with a little turpentine.

Finishing Varnishes

(*For white and coloured rings*).

These are made by triturating the various colours with the vehicle in a mortar. It will be found best, after preparing each colour, to place them in saucers such as are used by artists; the benzol then evaporates, and the colour can be taken up in a brush, as required, by simply moistening it with benzol.

The vehicle:—

Gum dammar	3 oz.
Gum mastic	1 oz.
Benzol	6 oz.

The colours:—

White	oxide of zinc.
Blue	ultramarine.
Red	carmine.
Black	lampblack.
Green	verdigris.
Yellow	chrome-yellow.

The foregoing formulæ are given in order that microscopists may, when desirable, be able to compound them for themselves. The author is of the opinion, however, that it will be found cheaper, where only small quantities are required, to purchase from a dealer in microscopical sundries, as most of these articles can only be *satisfactorily* made in large quantities. On the other hand, he would

also advise that the dealers in "secret nostrums" be not encouraged, for it is generally found that the greater the so-called secret, the more quackery there is about it, and the less utility.

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